

*Biodiversity  
and  
Animal Feed*

*Future Challenges  
for Grassland  
Production*

*Edited by*

**A. Hopkins  
T. Gustafsson  
J. Bertilsson  
G. Dalin  
N. Nilsson-Linde  
E. Spörndly**



**VOLUME 13  
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# *Biodiversity and Animal Feed*

## *Future Challenges for Grassland Production*

Proceedings of the 22<sup>nd</sup> General Meeting  
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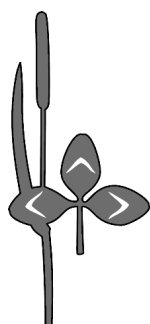
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## Foreword

As organisers of the 22nd EGF General Meeting, it is our great pleasure to welcome you to Uppsala on 9-12 June 2008. This Meeting is being organised by the Swedish University of Agricultural Sciences in cooperation with the Swedish Grassland Society. We are dedicating our best efforts to making this EGF 2008 conference memorable for all of you.

With the help of grasslands, it may be possible to combine safe and profitable food production with sustainable use of natural resources and preservation of biological and cultural values. The EGF 2008 conference will provide information about obstacles, conflicts of interest, new possibilities and good examples, many of them specifically related to the current severe global climate and food supply situation.

We are thrilled to see so many delegates participating in this most interesting conference. More than 450 participants from over 40 countries will attend the plenary, parallel and poster sessions, as well as master classes and technical excursions. Hopefully, the closing panel discussion on the theme 'Grasslands – Challenges for the Future' will also provide the audience with some challenges for future research and political decisions.

We are proud to present this proceedings volume, which contains up to 300 scientific reports. Covering a wide range of subjects on the theme of EGF 2008, *Biodiversity and animal feed – future challenges for grassland production*, these papers are of high scientific quality and information value and provide a great opportunity to update and expand our knowledge during the Uppsala meeting.

Active participation, intense discussions, new insights and new contacts are some of the features that we expect to characterise EGF 2008. However, perhaps most importantly of all, it is our hope that participants at this conference will ensure that EGF 2008 forms an important link in the chain of EGF Meetings.

On behalf of the organisers, we would like to express our gratitude to all those who have contributed to the EGF 2008 conference. Many people have unreservedly and enthusiastically participated in the planning and preparation of these four days in Uppsala, including the Organising and Scientific Committees, the speakers, the conference office, stewards, technicians, exhibitors, etc. Thank you all for your positive attitude and fantastic support! Thanks also to the supporters and sponsors of the meeting for their financial support, which made it possible to keep the registration fee for the individual participants at a low level.

Finally, special thanks to you, the delegates, for supplying the input needed for a successful scientific conference – high-quality papers and abstracts to be presented during plenary, parallel and poster sessions. We wholeheartedly welcome you and hope you find EGF 2008 as successful and rewarding as we envision it to be.

Göran Dalin  
President  
European Grassland Federation

Nilla Nilsson-Linde  
General Secretary  
Organising Committee EGF 2008



## Contents

<b>Opening Session. Biodiversity and animal feed .....</b>	<b>1</b>
<i>Invited papers</i>	
Semi-natural grasslands in Europe today .....	3
<i>Emanuelsson U.</i>	
Challenges for grasslands, grassland-based systems and their production potential in Europe .....	9
<i>Peeters, A.</i>	
 <b>Session 1. Biodiversity and productivity in grasslands – strategies and limitations .....</b>	 <b>25</b>
<i>Invited papers</i>	
Is the grass always greener on the other side of the fence? <i>Primula veris</i> L. as an example of plant survival at different management intensities .....	27
<i>Wissman J., Lennartsson T. and Berg, Å.</i>	
The benefits of sward diversity for cultivated grasslands .....	39
<i>Helgadóttir A., Connolly J., Collins R., Fothergill M., Kreuzer M., Lüscher A., Porqueddu C., Sebastià M.T., Wachendorf M., Brophy C., Finn J.A., Kirwan L. and Nyfeler D.</i>	
 <b>Session 1A. Enhanced plant and animal diversity in semi-natural grasslands .....</b>	 <b>53</b>
<i>Oral presentations</i>	
Effects of agri-environmental agreements on acridids and plant species richness in alfalfa crops .....	54
<i>Badenhausser I., Médiène S., Paris Le Clerc N. and Bretagnolle V.</i>	
Farm structure had only marginal effects on grassland diversity at a plot and farm level .....	57
<i>Weyermann I., Schneider M., Mosimann B., Hartmann G.A., Buholzer S. and Lüscher A.</i>	
Ecological rotation favours biodiversity in beef cattle systems in the French Massif Central .....	60
<i>Farruggia A., Dumont B., Leroy T., Duval C., and Garel J.-P.</i>	
Effects of plant litter on seedling establishment in semi-natural grasslands .....	63
<i>Hovstad K.A.</i>	
Troublesome plants in species-rich grasslands: can we maintain the toxic plant <i>Carex brevicollis</i> and alleviate its risk to livestock? .....	66
<i>Ruiz de los Mozos I., Oreja A., San Emeterio L., Zabalgogazcoa I. and Canals R.M.</i>	
Vegetation and butterfly diversity in a species-rich mountain pasture grazed by cattle under different stocking rates .....	69
<i>Dumont B., Farruggia A., Bachelard P., Frain M., Michaud Y. and Garel J.-P.</i>	
 <i>Poster presentations</i>	
Effect of fodder plantations of <i>Atriplex canescens</i> on the floristic diversity at nine stations in the department of Laghouat (Algeria) .....	72
<i>Amghar F., Arkoub A., Ben ahmed L., Ben saha K. and Ferrioune L.</i>	

Effects of agricultural practices on spatial dispersal patterns of permanent grassland species .....	75
<i>Amiaud B., Merlin A., Damour A., Gaujour E. and Plantureux S.</i>	
Effect of different grazing systems on communities of epigeic beetles in a submontane area .....	78
<i>Bohac J.J. and Slachta M.</i>	
Relationship between vascular plant species richness and soil chemical properties of alpine meadows and pastures .....	81
<i>Bohner A.</i>	
The effect of sowing date, cover and catch crops on the productivity of resown pasture swards .....	84
<i>Butkuvienė E.</i>	
Efficiency of environmental improvements in open areas of the Central Apennines Mountains (Italy) .....	87
<i>Cervasio F., Di Leo V., Ponzetta M.P., Argenti G., Genghini M. and Sacconi F.</i>	
Promoting biodiversity on intensively managed grassland in Scotland .....	90
<i>Cole L.J., Robertson D., Harrison W., McCracken D.I. and Tiley G.E.</i>	
A simplified method to determine the abundance of grass functional groups in natural grasslands .....	93
<i>Fallour D., Theau J.P., Corler K., Hossard L., Martin G., Jouany C., Duru M. and Cruz P.</i>	
Specific and functional diversity of vegetation in three beef cattle farms of the French Massif Central .....	96
<i>Farruggia A., Dumont B., Jouven M., Baumont R. and Loiseau P.</i>	
Effect of intensifying land use on community structure, functional diversity and productivity in calcareous grasslands .....	99
<i>Fayolle A., Lochet J., Hubert D., Cruz P., Navas M-L., Roumet C., Autran P., Jouany C. and Garnier E.</i>	
Tree density and fertilization regime effect on pasture biodiversity in a silvopastoral system established under <i>Pinus radiata</i> D. Don .....	102
<i>Fernandez-Nuñez E., Rigueiro-Rodriguez A. and Mosquera-Losada M.R.</i>	
Diet selection and herbage intake by cattle and sheep grazing two contrasting heathland communities .....	105
<i>Fraser M.D., Theobald V.J., Morris S.M. and Moorby J.M.</i>	
Performance of Limousin cross and Belted Galloway suckler cows and calves when grazing improved and semi-natural pasture .....	108
<i>Fraser M.D., Vale J.E. and Evans J.G.</i>	
Bird communities on Swedish wet meadows – importance of management regimes and landscape composition .....	111
<i>Gustafson T. and Berg Å.</i>	
Stability of the greenery in different light conditions in the old park at Falenty, Poland ....	114
<i>Gutkowska A., Burs W. and Wróbel B.</i>	
Foraging behaviour and diet composition in cattle on semi-natural grasslands .....	117
<i>Hessle A. and Wissman J.</i>	
Effects of social environment on grazing behaviour and liveweight changes in first-season-grazing dairy calves .....	120
<i>Hessle A. and Spörndly E.</i>	
The effect of grassland renovation and break up on nitrogen losses .....	123
<i>Kayser M., Seidel K., Müller J. and Isselstein J.</i>	

Influence of the site moisture and trophic conditions on the floristic diversity of meadow communities .....	126
<i>Kryszak A., Kryszak J. and Klarzynska A.</i>	
Effects of grassland restoration on plant species richness in Swedish agricultural landscapes .....	129
<i>Lindborg R. and Eriksson O.</i>	
Biodiversity and ecosystem services in mountain grasslands: a case study in the Planes de Son (Central Pyrenees) .....	132
<i>Llurba R., de Lamo X., Cuadros M., Ventura D. and Sebastià M.T.</i>	
Evolution of different pasture species in a silvopastoral system of NW Spain .....	135
<i>López-Díaz M.L., Mosquera-Losada M.R., and Rigueiro-Rodríguez A.</i>	
The effects of different kinds of livestock farming and abandonment on botanical diversity in mountain hay meadows .....	138
<i>López-i-Gelats F. and Bartolomé J.</i>	
Analysis of weed diversity in alfalfa ( <i>Medicago sativa</i> ) .....	141
<i>Médiène S.</i>	
Ecological and economical technology for mechanization of grassland improvement works .....	144
<i>Mocanu V., Hermenean I. and Marusca T.</i>	
Temporal distribution of cattle and horses grazing on mountain rangelands partially invaded by <i>Euphorbia polygalifolia</i> .....	147
<i>Mora M.J., Frutos P., Bedia J. and Busqué J.</i>	
Species rich limestone grasslands of the Burren, Ireland: feed value and sustainable grazing systems .....	150
<i>Moran J., Parr S., Dunford B. and O'Conchúir R.</i>	
Management strategies to restore agriculturally affected meadows on peat – biomass and N, P-balances .....	153
<i>Nielsen A.L. and Hald A.B.</i>	
<i>Betula pendula</i> , <i>Pinus sylvestris</i> and <i>Quercus robur</i> trees in Swedish semi-natural pastures – effects on plant diversity .....	156
<i>Pihlgren A. and Svensson R.</i>	
Management strategies to reduce regrowth species and increase biodiversity in semi-natural grasslands in central Norway .....	159
<i>Rosef L. and Bele B.</i>	
Analysis of the influence of some invasive plants species on the pastoral value of western Romanian grasslands .....	162
<i>Sărățeanu V., Moisuc A. and Butnariu M.</i>	
Biodiversity: the process from scientific results into farming practice .....	165
<i>Schiess C. and Schüpbach H.</i>	
An alternative paddock management system and its effect on grazing patterns and milk yield .....	168
<i>Schmitt, A., Murphy B. and Benedete H.</i>	
The seasonal biomass distribution of coprophagous beetles in fresh cow dung in a sub-mountain pasture .....	171
<i>Šlachta M., Frelich J. and Svoboda L.</i>	
Seed mixtures to establish species-rich meadows in the Swiss Central Plateau.....	174
<i>Suter D., Zanetti S. and Lüscher A.</i>	
Does the effect of selection history influence vegetation and plant preferences by cattle? .....	177
<i>Sæther N., Sickel H., Norderhaug A., Sickel M. and Vangen O.</i>	

Long-term effects of mulching on botanical composition, yield and nutrient budget of permanent grassland .....	180
<i>Tonn B. and Briemle G.</i>	
The influence of grassland management on biodiversity in the mountainous region of NE Romania .....	183
<i>Vintu V., Samuil C., Sarbu C., Saghin Gh. and Iacob T.</i>	
<b>Session 1B. Productivity in cultivated grasslands .....</b>	<b>187</b>
<b>Oral presentations</b>	
Simulating the morphogenesis and cutting of perennial ryegrass ( <i>Lolium perenne</i> ) plants with a 3D model .....	188
<i>Verdenal A., Combes D. and Escobar-Gutierrez A.</i>	
Sensor-based technologies to assess grassland biomass in short term leys .....	191
<i>Fricke T., Richter F. and Wachendorf M.</i>	
Productivity effects of grass-legume mixtures on two soil types .....	194
<i>Goliński P. and Golińska B.</i>	
Well-balanced grass-legume mixtures with low nitrogen fertilization can be as productive as highly fertilized grass monocultures .....	197
<i>Nyfelner D., Huguenin-Elie O., Suter M., Frossard E. and Lüscher A.</i>	
Herbs in grasslands – effect of slurry and grazing/cutting on species composition and nutritive value .....	200
<i>Søgaard K., Eriksen J. and Askegaard M.</i>	
Overwintering of timothy and perennial ryegrass in Norway from a climate change perspective .....	203
<i>Höglind M., Jørgensen M., Østrem L., Bakken A.K. and Thorsen S.M.</i>	
<b>Poster presentations</b>	
Yield potential, correlation and path-coefficient for two synthetics and three commercial varieties of alfalfa .....	206
<i>Abdel-Galil M.E.-D.M.</i>	
Yield potential and stability performance of some Egyptian clover genotypes under different environments .....	209
<i>Abdel-Galil M.E.-D.M., Helmy A.A., Sharawy W.M. and El-Nahrawy M.A.-Z.</i>	
Ability of grasses to compete in galega swards .....	212
<i>Baležentienė L. and Klimas E.</i>	
Pasture establishment from different seed mixtures of grass and clover species under varied seeding conditions .....	215
<i>Black A.D. and O'Kiely P.</i>	
Leaf functional traits in <i>Brachypodium pinnatum</i> (L.) Beauv., <i>Bromus erectus</i> Hudson and <i>Dactylis glomerata</i> L. as indicators of resource availability in grassland communities .....	218
<i>Bolzan A., Ferroni L., Vecchiettoni M. and Speranza M.</i>	
Effects of cutting management on the floristic composition of two permanent Italian ryegrass ( <i>Lolium multiflorum</i> ) meadows .....	221
<i>Bonifazzi B., Da Ronch F., Susan F. and Ziliotto U.</i>	
Study on long-term meadow productivity and botanical composition in response to different liming and fertilization .....	224
<i>Butkutė R. and Daugėlienė N.</i>	



Nitrogen fixation in clover grasslands of varied plant species richness .....	227
<i>Carlsson G., Palmborg C., Jumpponen A., Scherer-Lorenzen M., Högberg P. and Huss-Danell K.</i>	
Site effects on species productivity and dynamics: results from COST 852 in Wales .....	230
<i>Collins R.P., Fothergill M., Rees E., Kirwan L., Brophy C. and Connolly J.</i>	
Seasonal dynamics of dandelions ( <i>Taraxacum officinale</i> L.) in permanent grassland .....	233
<i>Daugeliene N. and Zekoniene V.</i>	
Potential of fodder legumes under intensive farming conditions in Flanders .....	236
<i>De Vlieghe A. and Carlier L.</i>	
Genotypic and symbiotic characters among rhizobia nodulating red clover in soils of northern Scandinavia .....	239
<i>Duodu S., Carlsson G., Huss-Danell K. and Svenning M.M.</i>	
Genetic characterization of indigenous root nodule bacteria from <i>Trifolium alexandrinum</i> L. cultivated on the island of Sardinia .....	242
<i>Floris R., Fois N., Sitzia M. and Salis L.</i>	
Comparison of yields and biological nitrogen fixation of two legumes grown in different silt soils .....	245
<i>Fustec J. and Bernard F.</i>	
Analysis of <i>Festulolium</i> and hybrid ryegrass ( <i>Lolium x boucheanum</i> ) dry matter yield stability .....	248
<i>Gutmane I. and Adamovich A.</i>	
The effect of root morphology on alfalfa yield in the spring regrowth period .....	251
<i>Hakl J., Santrucek J. and Krajic L.</i>	
Importance of polyploidy for forage and seed productivity and forage quality of perennial ryegrass .....	254
<i>Katova A., Ilieva A., Baert J., Hristov K. and Van Bockstaele E.</i>	
Genotype x environment influence on first-cut dry matter yield stability of annual ryegrass cultivars .....	257
<i>Kemešytė V. and Tarakanovas P.</i>	
The influence of management and exploitation of grasslands on the differentiation of their typological structure, biodiversity and productivity .....	260
<i>Klimeš F., Kobes M. and Suchý K.</i>	
The problems and perspectives of the grassland in Belarus .....	263
<i>Kulakouskaya T.V.</i>	
Yields of the sward depending on utilization method and species tested in mixtures .....	266
<i>Kulik M.A. and Warda M.</i>	
Grass and legume productivity oscillations in a binary mixture .....	269
<i>Lazaridou M.</i>	
Productivity and competitive ability evaluation of selected perennial ryegrass varieties, tested in different soil-climatic conditions .....	272
<i>Macháč R., Cagaš B., Pelikán J. and Hejduk S.</i>	
Sward regeneration after winter grazing depending on the intensity of sward damage .....	275
<i>Mattern T. and Laser H.</i>	
Weed flora dynamics during the first years of grassland establishment .....	278
<i>Médiène S. and Charrier X.</i>	
Forage yields in urban populations of hairy vetch ( <i>Vicia villosa</i> Roth) from Serbia .....	281
<i>Mihailović V., Mikić A., Vasiljević S., Katić S., Karagić Đ. and Čupina B.</i>	
Forage yields in urban populations of narrow-leaved vetch ( <i>Vicia sativa</i> subsp. <i>nigra</i> (L.) Ehrh.) from Serbia .....	284
<i>Mikić A., Mihailović V., Čupina B., Vasiljević S., Krstić Đ. and Milić D.</i>	



Effects of composted, pelletized and anaerobically digested sewage sludge on pasture production after sowing in a silvopastoral system .....	287
<i>Mosquera-Losada M.R., Santiago-Freijanes J.J., Fernández-Núñez E. and Rigueiro-Rodríguez A.</i>	
Changes in some morphological characteristics of reed canary-grass ( <i>Phalaroides arundinacea</i> L.) during primary growth .....	290
<i>Nagy G.</i>	
Winter survival, yield performance and forage quality of <i>Festulolium</i> cvs. for Norwegian farming .....	293
<i>Østrem L. and Larsen A.</i>	
Recovery of fertilizer N in grassland communities of different diversity and composition .....	296
<i>Palmborg C. and Huss-Danell K.</i>	
Nitrogen efficiency of farm manure on permanent grassland in mountainous regions .....	299
<i>Poetsch E.M. and Resch R.</i>	
Legume-cruciferae mixture as a multi-purpose crop in almond orchards in a semiarid climate .....	302
<i>Ramos M.E. and Robles A.B.</i>	
Improved estimation of mountain grassland production by using local input data in a crop model .....	305
<i>Ruget F., Godfroy M. and Plantureux S.</i>	
Influence of mineral fertilization on production and quality of forage in a Sardinian pasture .....	308
<i>Salis L. and Vargiu M.</i>	
Grassland productivity and consumption under different tree cover in the Aysén region, Patagonia, Chile .....	311
<i>Sánchez-Jardón L., Casado M.A., Del Pozo A., Ovalle C., Acosta B. and De Miguel J.M.</i>	
Are herbage yield and yield stability affected by plant species diversity in sown pasture mixtures? .....	314
<i>Sanderson M.A.</i>	
SDS-protein and isozyme markers of alfalfa genotypes and their offspring in response to drought and salinity stresses .....	317
<i>Sharawy W.M., Abdel-Tawab F.M., Fahmy, E.M., and Ibraheem, M.R.</i>	
Productivity and quality of perennial ryegrass ( <i>Lolium perenne</i> L.) in western Lithuania .....	320
<i>Skuodienė R. and Repšienė R.</i>	
Different management regimes of fodder galega ( <i>Galega orientalis</i> ) sward for organic farming .....	323
<i>Slepetys J.</i>	
Production and morphological traits of tall oatgrass breeding families .....	326
<i>Sokolovic D., Radovic J., Lugic Z., Babic S. and Simic A.</i>	
Manipulating pasture grass growth by nitrogen fertilization .....	329
<i>Thomet P., Stettler M. and Hadorn M.</i>	
Influence of site conditions on interspecific interactions and yield of grass-legume mixtures .....	332
<i>Thumm U.</i>	
Leguminous species in ass. <i>Agrostietum vulgaris</i> and ass. <i>Festucetum valesiacae</i> on Stara Planina mountain .....	335
<i>Tomic Z., Nesic Z.D., Krnjaja V.S. and Zujovic M.M.</i>	

Radiation use and uptake of mineral N in leys under climatic change .....	338
<i>Torssell B., Eckersten H., Kornher A. and Nyman P.</i>	
Influence of root rot on the sustainability of grass/legume leys in Sweden .....	341
<i>Wallenhammar A.-C., Nilsdotter-Linde N., Jansson J., Stoltz E. and L-Baeckström G.</i>	
Persistency of <i>Lolium perenne</i> in sward under 10-year pasture utilization at varied wetness of peat-muck soil .....	344
<i>Warda M., Stamirowska-Krzaczek E.A. and Kulik M.A.</i>	
Effect of N fertilization on the development of couch-grass ( <i>Elytrigia repens</i> ) in a grass sward .....	347
<i>Varekova P., Svozilova M., Micova P. and Rzonca J.</i>	
The productivity of early type red clover ( <i>Trifolium pratense</i> ) varieties and breeding lines .....	350
<i>Vilčinskas E. and Sprainaitis A.</i>	
Performance of silage maize in forage crop rotation systems .....	353
<i>Wulfes R. and Ott H.</i>	
Changes in tillering and leaf chemistry in <i>Brachypodium phoenicoides</i> infected by the fungus <i>Epichloë typhina</i> .....	356
<i>Zabalgogezcoa I., García Ciudad A., Leuchtmann A., Vázquez de Aldana B.R. and García Criado B.</i>	
<b>Session 2. Grassland as part of the food chain .....</b>	<b>359</b>
<b><i>Invited papers</i></b>	
Botanically diverse forage-based rations for cattle: implications for product composition, product quality and consumer health .....	361
<i>Moloney A.P., Fievez V., Martin B., Nute G.R. and Richardson R.I.</i>	
Plant diversity in grasslands and feed quality .....	375
<i>Huyghe C., Baumont R. and Isselstein J.</i>	
<b>Session 2A. Food quality .....</b>	<b>387</b>
<b><i>Oral presentations</i></b>	
Effects of the grazed horizon in perennial ryegrass swards on the conjugated linoleic acid concentration in milk of dairy cows .....	388
<i>Elgersma A., Van der Hoeven E., Witkowska I. and Smit H.J.</i>	
Effects of harvest time of red clover silage on milk production and composition .....	391
<i>Vanhatalo A., Pursiainen P., Kuoppala K., Rinne M. and Tuori M.</i>	
Influence of botanically different forages on rumen, milk and tissue fatty acid metabolism .....	394
<i>Lourenço M., Van Ranst G., De Smet S. and Fievez V.</i>	
Growth and carcass quality of suckler calves grazing at the farm or in the mountains .....	397
<i>Steinshamn H., Höglind M., Havrevoll Ø., Saarem K., Lombnæs I.H. and Svendsen A.</i>	
Volatile compounds of Alpine vegetation as markers for the traceability of 'grass-deriving' dairy products .....	400
<i>Lombardi G., Falchero L., Coppa M. and Tava A.</i>	

### **Poster presentations**

Organic selenium providing by selenized fertilization in grazed grass, grass and maize silage for beef and dairy cows .....	403
<i>Cabaraux J.-F., Hornick J.-L., Dotreppe O., Istasse L., Paeffgen S. and Dufrasne I.</i>	
No effects on milk fatty acid composition of perennial ryegrass cultivars with contrasting forage quality .....	406
<i>Elgersma A., Van der Horst H., Dekker R., Ellen G. and Tamminga S.</i>	
Lamb meat – effect of finishing system on quality .....	409
<i>Lind V., Jørgensen M., Mølmann J., Haugland E. and Hersleth M.</i>	
Effect of different grazing times on milk fatty acid composition .....	412
<i>Morales-Almaraz E., Vicente F., Soldado A., Martínez-Fernández A. and de la Roza-Delgado B.</i>	
Effects of vegetation and grazing preferences on the quality of alpine dairy products .....	415
<i>Sickel H., Abrahamsen R., Lunnan T., Norderhaug A. and Ohlson M.</i>	
Somatic cell count and quality of milk during pasture turnout of dairy cows .....	418
<i>Wredle E., Östensson K. and Svennersten-Sjaunja K.</i>	
Influence of forage from grassland on the fatty acid content of milk fat .....	421
<i>Wyss U. and Collomb M.</i>	

### **Session 2B. Feed quality ..... 425**

#### **Oral presentations**

Estimation of nutritive value of grasses from semi-natural grasslands by biological, chemical and enzymatic methods .....	426
<i>Aufrère J., Carrère P., Dudilieu M. and Baumont R.</i>	
Effects of seed mixture and N fertilization on nitrate content of grass-legume swards .....	429
<i>Nešić Z., Tomić Z., Ružić-Muslić D. and Vučković S.</i>	
Contents of $\alpha$ -tocopherol and $\beta$ -carotene in grasses and legumes harvested at different maturities .....	432
<i>Danielsson H., Nadeau E., Gustavsson A.-M., Jensen S.K., Søgaard K. and Nilsdotter-Linde N.</i>	
Effect of plant mediated proteolysis on protein degradation among ten diploid perennial ryegrass ( <i>Lolium perenne</i> L.) genotypes .....	435
<i>Lösche M., Salama H., Gierus M., Herrmann A., Voss P. and Taube F.</i>	
Agronomic potential of tanniferous forage plant species and cultivars .....	438
<i>Häring D.A., Suter D. and Lüscher A.</i>	

### **Poster presentations**

The effect of nitrogen fertilizer and harvesting frequency on yield and <i>in-vitro</i> digestibility of barley grass .....	441
<i>Asadi E., Moharrery A. and Hvelplund T.</i>	
Quality development in regrowths of timothy, meadow fescue and red clover .....	444
<i>Bakken A.K., Lunnan T. and Höglind M.</i>	
<i>In-vitro</i> gas production of forage from semi-natural grassland as influenced by N fertilization .....	447
<i>Bošnjak K., Knežević M., Leto J., Vranić M., Perčulija G., Matić I. and Kutnjak H.</i>	
Stability of fatty acids in grass silages after exposure to air .....	450
<i>Cone J.W., Meulenberg S., Elgersma A. and Hendriks W.H.</i>	

Preservatives and machines with knives improve the silage quality in round bales .....	453
<i>Eriksson H.</i>	
Digestibility and fibre content of leaves and straw of three <i>Festulolium</i> hybrids during spring regrowth .....	456
<i>Frankow-Lindberg B.E. and Olsson K.-F.</i>	
Quality of forage maize ( <i>Zea mays</i> L.) depending on sowing time in Latvia .....	459
<i>Gaile Z.</i>	
Changes in feed quality of different forage legumes during autumn growth .....	462
<i>Gierus M., Kleen J. and Taube F.</i>	
Changes in water soluble carbohydrates for perennial ryegrass in the first growth in Norway and Sweden .....	465
<i>Halling M.A. and Nesheim L.</i>	
Endophytic fungi ( <i>Neotyphodium</i> ) in grasses. What is the situation in Sweden? .....	468
<i>Huss-Danell K.</i>	
Quality of timothy ( <i>Phleum pratense</i> L.) and causality of its variation .....	471
<i>Jonavičienė K., Paplauskienė V., Lemežienė N. and Butkutė B.</i>	
Contents of phenolics, protein precipitation capacity of tannin in browse and ability to predict them in diets from faecal composition in sheep .....	474
<i>Kumara Mahipala M.B.P., Krebs G.L., McCafferty P. and Dods K.</i>	
Improvement of quality of silage made of bird's foot trefoil by using additives and bird's foot-timothy mixture .....	477
<i>Lättemäe P. and Tamm U.</i>	
Nutritive value in leaves and stems of lucerne with advanced maturity and a comparison of methods for determination of lignin content .....	480
<i>Markovic J., Radovic J., Lugic Z. and Sokolovic D.</i>	
The effect of botanical composition on mineral content in hay from extensively managed mountain grasslands .....	483
<i>Misztal A. and Zarzycki J.</i>	
Characterization of water soluble proteins in some forage species and their effects on fermentation process measured by a gas production method .....	486
<i>Moharrery A. and Hvelplund T.</i>	
The importance of timothy variety for feed quality development in regrowths of swards mixed with meadow fescue .....	489
<i>Nesheim L. and Bakken A.K.</i>	
Red clover for silage: management impacts on chemical composition in the season after sowing .....	492
<i>O'Kiely P. and Black A.D.</i>	
Effect of different tiller types on the accumulation and digestibility of the herbage mass of timothy ( <i>Phleum pratense</i> L.) .....	495
<i>Pakarinen K., Virkajärvi P., Seppänen M. and Rinne M.</i>	
Forage quality by animal fertilizer applications and by different grassland management .....	498
<i>Pozdisek J., Stybnarova M., Kohoutek A., Svozilova M. and Rzonca J.</i>	
Classification of mountain permanent grasslands based on their feed value .....	501
<i>Rodrigues A. M., Andueza D., Picard F., Cecato U., Farruggia A. and Baumont R.</i>	
Digestibility and fibre fractions of perennial ryegrass ( <i>Lolium perenne</i> L.) as affected by cultivar and ploidy level .....	504
<i>Salama H., Lösche M., Herrmann A., Gierus M. and Taube F.</i>	

Changes in the non-structural carbohydrate content of two cool-season steppe grasses under different cutting frequencies .....	507
<i>Schiborra A., Gierus M., Wan H., Bai Y. and Taube F.</i>	
Fibre fractions of red clover ( <i>Trifolium pratense</i> L.) at different harvests over two seasons .....	510
<i>Vasiljevic S., Glamocic D., Jajic I., Cupina B., Katic S., Milic D. and Mikic V.</i>	
Feeding value of legumes and grasses at different harvest times .....	513
<i>Weisbjerg M.R. and Søgaard K.</i>	
<b>Session 3. Efficient resource utilisation for sustainable conventional and organic grassland production .....</b>	<b>517</b>
<b><i>Invited papers</i></b>	
Sustainable nutrient management for organic farming systems .....	519
<i>Watson C.A. and Wachendorf M.</i>	
Recent developments in harvesting and conservation technology for feed and biomass production of perennial forage crops .....	529
<i>Orosz S., Szűcsné-Péter J., Owens V. and Bellus Z.</i>	
<b>Session 3A. Organic production and nutrient flows .....</b>	<b>549</b>
<b><i>Oral presentations</i></b>	
Towards nitrogen self-sufficiency in mixed crop organic dairy systems: legumes and protein-rich plant contributions .....	550
<i>Coquil X., Foissy D., Trommenschlager J.M., Bazard C., Delaby L. and Despres S.</i>	
Effects of different stocking rates with dairy cows on herbage quality in organic farming .....	553
<i>Schori F.</i>	
Productivity and N-leaching in organic dairy grass-arable crop rotations .....	556
<i>Eriksen J., Askegaard M. and Søgaard K.</i>	
In-field N transfer, build-up, and leaching in ryegrass-clover mixtures .....	559
<i>Rasmussen J., Eriksen J., Jensen E.S. and Høgh-Jensen H.</i>	
Nitrogen fixation and nitrogen allocation in red clover-grasslands .....	562
<i>Huss-Danell K., Chaia E. and Carlsson G.</i>	
<b><i>Poster presentations</i></b>	
Hay and silage as vitamin sources in organic sheep production .....	565
<i>Bernes G., Persson Waller K. and Jensen S.K.</i>	
Analysis of hyperspectral data to estimate dry matter yield of legume-grass swards .....	568
<i>Biewer S., Fricke T. and Wachendorf M.</i>	
Influence of organic fertilizers on the dry matter yield and microelement content of meadow herbage .....	571
<i>Ciepiela A.G., Kolczarek R., Jankowska J., Jodelka J. and Jankowski K.</i>	
Establishing a risk index to prevent nitrogen non-point pollution of waters in the Basque Country .....	574
<i>Del Hierro O., Artetxe A., De Francisco M. and Pinto M.</i>	

Milk urea content: effects of environmental parameters and relationships with other milk traits .....	577
<i>Dufresne I., Cabaraux J.-F., Istasse L. and Hornick J.-L.</i>	
Strategy of organic fertilizer use on permanent grassland – results of a 22-year-old experiment on meadow and mowing-pasture .....	580
<i>Elsaesser M., Kunz H.G. and Briemle G.</i>	
Apparent balance of nitrogen in organic and conventional dairy farms in Tuscany (Central Italy) .....	583
<i>Giustini L., Argenti G. and Acciaioli A.</i>	
Effect of bio and mineral fertilization on fresh weight and dry matter forage yields of three pearl millet varieties .....	586
<i>Helmy A.A., Abdel-Galil M.M., Abdel-Gawad M.S. and Sharawy W.M.</i>	
The application of image analysis to estimate legume contents in legume / grass swards .....	589
<i>Himstedt M., Wachendorf M. and Fricke T.</i>	
Balancing milk yield and stocking rate to the output from an organic crop rotation system .....	592
<i>Johansen A., Bakken A.K. and Hansen S.</i>	
Effects of intensity of fertilization and cutting frequency on forage quality and diversity of permanent grassland in Central Europe in 2003-2007 .....	595
<i>Kohoutek A., Komárek P., Nerušil P., Odstrčilová V., Hrabě F., Rosická L., Šrámek P., Kašparová J., Gaisler J., Fiala J., Pozdíšek J., Míčová P., Svozilová M. and Jakešová H.</i>	
Effects of summer sown catch crops on soil N <sub>min</sub> and N removal with herbage before winter .....	598
<i>Kramberger B. and Gselman A.</i>	
Long-term changes of <sup>15</sup> N natural abundance of plants and soil in grassland .....	601
<i>Kriszan M., Kühbauch W., Amelung W., Schellberg J. and Gebbing T.</i>	
Alternative feeding regimes to ewes in organic farming in northern Norway .....	604
<i>Lind V. and Eilertsen S.M.</i>	
Effect of different management systems on sown meadows .....	607
<i>Martínez-Fernández A., Pedrol N., Martínez A., Soldado A., Vicente F. and de la Roza-Delgado B.</i>	
The effect of cattle-slurry electroflotation on gaseous emissions from grasslands .....	610
<i>Menéndez S., Merino P., Pinto M., Lekuona A., González-Murua C. and Estavillo J.M.</i>	
Effects of direct seeding and cow slurry application on maize production in fine-textured soils .....	613
<i>Mijangos I., Albizu I., Epelde L., Pinto M., del Hierro O., Arriaga H. and Garbisu C.</i>	
Algae residue to enhance pasture production .....	616
<i>Mosquera-Losada M.R., Fernández-Núñez E., Santiago-Freijanes J.J. and Rigueiro-Rodríguez A.</i>	
Productive and economic results for organic farms with a large share of grassland in Poland in the years 2004-2006 .....	619
<i>Prokopowicz J. and Jankowska-Huflejt H.</i>	
The effects of red clover on winter wheat yields .....	622
<i>Repsiene R. and Skuodiene R.</i>	
The impact of legumes on cereals in a crop rotation and legume-cereal bi-cropping .....	625
<i>Sarunaite L., Kadziulienė Z. and Kadziulis L.</i>	



Seasonality of productivity, botanical composition and N concentrations of four forage legume-grass mixtures under cutting .....	628
<i>Smit H.J., Nepal S., Van Vilsteren D., Witkowska I.M. and Elgersma A.</i>	
Evaluation of the contribution of soil organic matter mineralization to harvested N in temperate grasslands .....	631
<i>Stroia C., Cruz P., Duru M., Justes E., Martin G., Theau J.P. and Jouany C.</i>	
Inter-annual variations in phosphorus content of semiarid grasslands over a long time period .....	634
<i>Vázquez-de-Aldana B.R., García-Ciudad A., Petisco C. and Garcia-Criado B.</i>	
The influence of organic and mineral fertilizers on fodder quality in NE Romania .....	637
<i>Vintu V., Samuil C., Trofin A. and Popovici I.C.</i>	
<b>Session 3B. Forage conservation .....</b>	<b>641</b>
<b>Oral presentations</b>	
The effect of silage making technology on production and quality of milk .....	642
<i>Jaakkola S., Saarisalo E., Heikkilä T., Nysand M., Suokannas A., Mäki M. and Taimisto A.M.</i>	
Quality and economics of pre-wilted silage made by wide-spreading or by swathing .....	645
<i>Spörndly R., Knicky M., Pauly T. and Lingvall P.</i>	
Loader wagon compared to metered chopper for forage harvest .....	648
<i>Suokannas A. and Nysand M.</i>	
Quality and aerobic stability of big-bale silage treated with bacterial inoculants containing <i>Lactobacillus buchneri</i> .....	651
<i>Wróbel B.</i>	
Effect of conservation method on fatty acid composition in model grass silage .....	654
<i>Arvidsson K., Gustavsson A.-M. and Martinsson K.</i>	
<b>Poster presentations</b>	
Fatty acids in fresh and artificially dried grass .....	657
<i>Elgersma A. and Wever A.C.</i>	
Effects of climate and cutting delays on timeliness losses in silage harvest .....	660
<i>Gunnarsson C., Rosenqvist H., Spörndly R., Sundberg M. and Hansson P.-A.</i>	
Effects of a multi-strain inoculant on the fermentation characteristics of lucerne silage .....	663
<i>Jatkauskas J., Vrotniakiene V. and Ohlsson C.</i>	
Precision chopping or rotor cutting and its influence on ensiling capacity and silage fermentation .....	666
<i>Lingvall P. and Knicky M.</i>	
$\alpha$ -tocopherol and $\beta$ -carotene in baled silage and haylage .....	669
<i>Müller C., Möller J., Jensen S.K. and Udén P.</i>	
Storage of small bale silage and haylage .....	672
<i>Müller C., Pauly T. and Udén P.</i>	
Effects of slurry application and wide-spreading of forage on silage quality .....	675
<i>Nysand M., Suokannas A., Saarisalo E., Heikkilä T., Jauhiainen L., Taimisto A.-M. and Jaakkola S.</i>	
Inoculation of experimental silages with different <i>Clostridium</i> spores .....	678
<i>Pauly T., De Paula Sousa D., Spörndly R. and Christiansson A.</i>	

Handling round bale silage after stretch-film application .....	681
<i>Spörndly R., Nylund R., Hörndahl T. and Algerbo P.</i>	
Influence of wilting and ensiling ryegrass and clover with different additives on lipid metabolism .....	684
<i>Van Ranst G., Fievez V., De Riek J. and Van Bockstaele E.</i>	
Effects of multiple strains in an inoculant on the fermentation quality and nutritive value of grass-legume silage .....	687
<i>Vrotniakienė V. and Jatkauskas J.</i>	
 <b>Session 4. Can grassland-based production survive in Europe?</b>	
– pathways for future success .....	691
<b>Invited papers</b>	
Diversity of beef farming systems and grasslands use in Europe .....	693
<i>Sarzeaud P., Pflimlin A., Perrot C. and Becherel F.</i>	
To graze or not to graze, that's the question .....	706
<i>Van den Pol-van Dasselaar A., Vellinga T.V., Johansen A. and Kennedy E.</i>	
 <b>Session 4A. The future of the grassland farmer .....</b>	<b>717</b>
<b>Oral presentations</b>	
Perspectives in dairy production for family farms in different mountain regions of Switzerland .....	718
<i>Durgiai B. and Blättler T.</i>	
Mobile milking robot offers new grazing concept .....	721
<i>Oudshoorn F.W.</i>	
Local participation in seminatural grasslands maintenance: lessons from Sweden .....	724
<i>Stenseke M.</i>	
Biogas yields from grassland .....	727
<i>Prochnow A., Heiermann M., Idler C., Linke B., Plöchl M., Amon T., Langeveld H. and Hobbs P.</i>	
The boom in biomass production – a challenge for grassland biodiversity? .....	730
<i>Stein S. and Krug A.</i>	
Agroforestry systems: from tradition to future sustainable land use .....	733
<i>Mosquera-Losada M., Santiago-Freijanes J., Fernández-Núñez E. and Rigueiro-Rodríguez A.</i>	
 <b>Poster presentations</b>	
Home-grown forage use on commercial dairy farms in The Basque Country .....	736
<i>Arriaga H., Pinto M., Calsamiglia S. and Merino P.</i>	
Citizens' evaluation of rangeland resources for a sustainable regional development .....	739
<i>Arabatzi G., Kyriazopoulos A. and Lazaridou M.</i>	
Efficient utilization of grassland biomass for energy purposes through mechanical dehydration of silages .....	742
<i>Graß R., Reulein J. and Wachendorf M.</i>	
Impact of global climate change scenarios on alfalfa production in France .....	745
<i>Ruget F., Abdessemed A. and Moreau J.-C.</i>	
Perspectives of precision agriculture on grassland .....	748
<i>Schellberg J.</i>	



Suitability of low-intensity grassland for combustion as influenced by grassland community and harvest date .....	751
<i>Tonn B., Thumm U. and Claupein W.</i>	

#### **Session 4B. Forage-based animal production ..... 755**

##### ***Oral presentations***

Effect of pre-grazing herbage mass on production performance of dairy cows in mid-lactation .....	756
<i>McEvoy M., O'Donovan M., Boland T. and Delaby L.</i>	
Effect and carryover effect of spring grazing access time on dairy cow performance .....	759
<i>Delaby L., Peyraud J.L., Pérez-Ramírez E. and Delagarde R.</i>	
A new Nordic structure evaluation system for diets fed to dairy cows .....	762
<i>Nørgaard P., Nadeau E., Volden H., Randby Å., Aaes O. and Mehlqvist M.</i>	
Estimation of the digestion rate and digestibility of forages using <i>in-vitro</i> gas production technique .....	765
<i>Stefanska J., Rinne M., Seppälä A., Vanhatalo A., Nousiainen J. and Huhtanen P.</i>	
Effect of grass silage chop length on intake and milk production by dairy cows .....	768
<i>Randby Å.T., Garmo T., Eknæs M. and Prestløkken E.</i>	
The impact of cow genotype on the profitability of grassland-based beef production in Ireland .....	771
<i>Crosson P.</i>	

##### ***Poster presentations***

Kura clover and reed canarygrass: a versatile mixture for dairy production in northern environments .....	774
<i>Albrecht K.A., Combs D.K., Kammer K.L. and Gregoret R.</i>	
Evaluation of galega suitability for cattle feeding .....	777
<i>Balezentiene L.</i>	
The effects of forage type and maturity on digesta kinetics of large and small particles in dairy cows .....	780
<i>Bayat A.R., Rinne M., Kuoppala K., Ahvenjärvi S. and Huhtanen P.</i>	
Effects of chop length of silage on chewing time, feed consumption and milk production .....	783
<i>Bertilsson J.</i>	
Performance of continental weanling heifers grazing pasture or consuming grass silage during winter in Ireland .....	786
<i>Black A.D. and Moloney A.P.</i>	
Impacts of compact calvings and once-a-day milking in grassland based systems .....	789
<i>Brocard V. and Portier B.</i>	
Maintaining grassland-based production in regions with low rainfall and dry soils: Controlled Maine-Anjou Origin Appellation (AOC Maine-Anjou) .....	792
<i>Couvreux S., Laurent C., Schmitt T., Audic C. and Lautrou Y.</i>	
Ruminal fermentation pattern in dairy cows as affected by herbage proportion in the diet and feeding level .....	795
<i>Delagarde R., Pérez-Ramírez E. and Peyraud J.L.</i>	
Short time eating rate by goats and cows of long and chopped grass silage, harvested at three stages of growth .....	798
<i>Dønnem I., Eknæs M., Randby Å.T. and Garmo T.H.</i>	

Farmers' perspectives for forage-based dairy production. A Review .....	801
<i>Elgersma A.</i>	
Triticale and mixtures silages for feeding dairy cows .....	804
<i>Emile J.-C., Jacobs dias F., Al Rifāi M., Le Roy P. and Faverdin P.</i>	
The effectiveness of grazing management in terms of milk production on sub-mountain dairy farms .....	807
<i>Frelich J., Šlachta M., Čermák B. and Vávrová L.</i>	
Effect of grass silage chop length on chewing activity and digestibility .....	810
<i>Garmo T.H., Randby Å.T., Eknæs M., Prestløkken E. and Nørgaard P.</i>	
The effects of grazing pressure and concentrate use on efficient dairy production in Galicia .....	813
<i>Gonzalez-Rodriguez A., Vazquez O.P. and Lopez J.</i>	
Horse grazing: practices in Franche-Comté region of France .....	816
<i>Granger S., De Broca M., Dupuy D'Uby M., Marcheron H., Marsot M. and Roux M.</i>	
Voluntary intake of silage in small and large ruminants depending on chemical composition and <i>in-vitro</i> gas production characteristics .....	819
<i>Hetta M., Bernes G., Martinsson K., Krizsan S.J. and Randby Å.T.</i>	
Faecal particle-size distribution from ewes fed grass silages harvested at different stages of maturity .....	822
<i>Jalali A., Nørgaard P., Nadeau E., Arnesson A., Weisbjerg M.R.</i>	
Organic milk production based entirely on grassland feeds .....	825
<i>Johansson B. and Holtenius K.</i>	
Reverting to grazing: farmers' conceptions .....	828
<i>Michaud A., Havet A. and Mathieu A.</i>	
Previous nutrition and entry time to pasture affect weight gain and grazing behaviour in steers .....	831
<i>Milano G.D., Sánchez Chopa F., Nadin L.B. and Gonda H.L.</i>	
Performance of pregnant and lactating ewes fed grass silages differing in maturity .....	834
<i>Nadeau E. and Arnesson A.</i>	
Influence of silage structure on feeding behaviour and abnormal behaviours in dairy heifers .....	837
<i>Nielsen P., Nadeau E., Gustavsson M., Lidback F. and Lidfors L.</i>	
Milk production systems to increase competitiveness in regions of high rainfall and heavy clay soil types .....	840
<i>O'Donovan M., O'Loughlin J. and Kelly F.</i>	
Are dairy cow rumen pH levels affected by herbage mass and daily herbage allowance? .....	843
<i>Palladino R.A., O'Donovan M., Kenny D.A. and Mc Evoy M.</i>	
Effect of harvesting time and wilting of silage on digestibility in cows and sheep .....	846
<i>Prestløkken E., Randby Å.T., Eknæs M. and Garmo T.H.</i>	
Effect of harvesting time and wilting on feed intake and milk production .....	849
<i>Prestløkken E., Randby Å.T., Eknæs M. and Garmo T.H.</i>	
Performance of dairy steers fed whole-crop barley silages harvested at different stages of maturity .....	852
<i>Rustas B-O., Nadeau E. and Johnsson S.</i>	
Nitrogen excretion and utilization in grassland dairy cows .....	855
<i>Salcedo G. and Martinez-Suller L.</i>	

Evaluation of a dairy sheep system in a Sardinian hill area based on natural pasture: milk production and feedstuff supplementation .....	858
<i>Sitzia M. and Fois N.</i>	
Effect of mixed grazing of suckler cows and lambs on faecal egg counts and animal performance .....	861
<i>Sormunen-Cristian R., Manninen M., Jauhiainen L. and Oksanen A.</i>	
What type of cow do we need for grassland based milk production? .....	864
<i>Thomet P. and Kunz P.</i>	
Feed intake and digestibility of whole-crop cereal silages fed to dairy heifers .....	867
<i>Wallsten J., Nadeau E., Bertilsson J. and Martinsson K.</i>	
The effect of intake of total and digestible fibre on milk fat production in cows .....	870
<i>Wallsten J. and Martinsson K.</i>	
Influence of different participation of forages from grasslands in feeding ratio on effectiveness of dairy cattle feeding .....	873
<i>Wasilewski Z., Barszczewski J. and Wróbel B.</i>	
Effect of silage maize hybrid (dry down vs. stay green) on dairy cow performance .....	876
<i>Zom R.L.G., Van Schooten H.A. and Van Laar H.</i>	

## **Session 5. The grassland landscape as a base for animal production – present, past and future ..... 879**

### ***Invited papers***

The historical legacy on grassland plant diversity .....	881
<i>Cousins S.A.O.</i>	
Low-intensity livestock systems in Europe: an opportunity for quality products, recreation revenues and environmental conservation .....	892
<i>Sebastià M.T., Canals R.M., Marks E. and Llurba R.</i>	

## **Session 5A. The grassland landscape in Europe ..... 903**

### ***Oral presentations***

Historical grazing pressure in south-central Sweden, 1620-1850 .....	904
<i>Dahlström A.K.M.</i>	
Eighteenth century land use – a path to future conservation of grassland plant diversity .....	907
<i>Gustavsson E., Lennartsson T. and Emanuelsson M.</i>	
Agricultural grasslands in Bavaria – interrelationship of diversity and management .....	910
<i>Heinz S., Mayer F. and Kuhn G.</i>	
Effects of summer drought on temperate grassland performance .....	913
<i>Gilgen A.K. and Buchmann N.</i>	

### ***Poster presentations***

Botanical composition of mat-grass ( <i>Nardus stricta</i> ) grassland communities .....	916
<i>Alibegovic-Grbic S., Bezdrob M. and Murtic S.</i>	
Environmental characteristics of abandoned grasslands, production orientation and socio-economic types of farms in the pre-alpine region of Slovenia .....	919
<i>Borec A., Neve N. and Flambard A.</i>	
Dry grasslands of Switzerland: an overview .....	922
<i>Hedinger C. and Gubser C.</i>	

Farmland bird indicators for evaluation of habitat quality in agricultural landscapes dominated by grassland areas .....	925
<i>Hoffmann J., Kiesel J. and Greef J.-M.</i>	
Economic evaluation of the grasslands in two regional parks in Italy .....	928
<i>Marzetti S., Disegna M., Villani G. and Speranza M.</i>	
Variations in the phenology of semi-natural meadows in the western part of Switzerland .....	931
<i>Meisser M., Amaudruz M. and Jeangros B.</i>	
Landscape values of grasslands .....	934
<i>Nagy G. and Wiwczarowski T.</i>	
Influence of some climatic factors on invasive species coverage index in western Romania .....	937
<i>Sărățeanu V., Moisuc A., Oriol I. and Butnariu M.</i>	
Prediction of the potential effects of increased flooding periods on grassland in the core area of the German-Polish National Park .....	940
<i>Schalitz G., Behrendt A. and Rogge H.</i>	
Long-term effects of N, P and K fertilization on specific biodiversity in a permanent mountain meadow .....	943
<i>Susan F. and Ziliotto U.</i>	
Botanical and ecological description of pasture types in an area of the Italian Alps .....	946
<i>Targetti S., Staglianò N., Argenti G. and Messeri A.</i>	
Feral sheep in coastal heaths – developing sustainable agriculture in vulnerable cultural landscapes .....	949
<i>Velle L.G., Norderhaug A. and Øpstad S.L.</i>	
The botanical changes of the <i>Lolio-Cynosuretum</i> under different management conditions .....	952
<i>Vozár L., Jančovič J. and Fillo M.</i>	
Photoperiodic effects on elongation growth of two timothy ecotypes .....	955
<i>Wu Z., Baadshaug O.H. and Skjelvåg A.O.</i>	
Floristic diversity and bioindicative assessment of the meadow habitats in the Huczwa river valley .....	958
<i>Wyłupek T., Harkot W. and Lipińska H.</i>	
Floristic diversity and economic value of permanent grasslands in the Por river valley .....	961
<i>Wyłupek T.</i>	
The effect of environmental factors on the occurrence of legumes in mountain grasslands .....	964
<i>Zarzycki J. and Misztal A.</i>	

## **Session 5B. Low-intensity systems in future grassland production**

### **– benefits and risks ..... 967**

#### **Oral presentations**

Performance of beef suckler cows and their progeny to slaughter on intensive and extensive grassland systems .....	968
<i>McGee M. and Drennan M.J.</i>	
The effect of outdoor cattle keeping in winter season on water and soil quality .....	971
<i>Svozilova M., Rzonca J. and Varekova P.</i>	

Simulating the impact of dry climatic years on grassland utilization and production from beef cattle farming systems based on permanent pasture .....	974
<i>Baumont R., Deux N., Farruggia A. and Jouven M.</i>	
Impact of mixed grazing on animal performance and moorland vegetation .....	977
<i>McLean B.M., Davies O.D., Critchley N., Griffiths B., Adamson H. and Evans E.</i>	
<b>Poster presentations</b>	
Evaluating an organic low input grassland dairy system farming on permanent pastures in eastern France (Vosges lowland) .....	980
<i>Fiorelli J.-L., Coquil X., Gouttenoire L., Gaujour E., Bazard C., Trommenschlager J.-M. and Mignolet C.</i>	
Economy of beef production on extensive pasture in a nature conservation area .....	983
<i>Goliński P., Golińska B., Biniaś J. and Zajac M.</i>	
The nutritional value of pasture forage for sheep in the Krkonoše Mountains National Park .....	986
<i>Homolka P.</i>	
Effects of natural pasture on body condition in fit Standardbred geldings .....	989
<i>Jansson A.</i>	
Long term studies to determine management practices to enhance biodiversity within semi-natural grassland communities .....	992
<i>Morgan M., McLean B.M. and Davies O.D.</i>	
Limited utilization impact on productivity and floristic diversity of grasslands in the Sudeten mountains .....	995
<i>Nadolna L., Fatyga J., Zyszkowska M. and Paszkiewicz-Jasinska A.</i>	
Crop and ley-rotation challenges for low input dairy farming systems – a planned farm study .....	998
<i>Parente G., Altobelli A. and Dovier S.</i>	
Vegetation type selected by cattle grazing heterogeneous semi-natural pastures .....	1001
<i>Pelve M.E. and Spörndly E.</i>	
Effect of shading on biodiversity of cocksfoot ( <i>Dactylis Glomerata</i> ) established sward .....	1004
<i>Rodriguez-Barreira S., Mosquera-Losada M.R. and Rigueiro-Rodriguez A.</i>	
Weight gain among steers grazing semi-natural pastures every other year compared with every year .....	1007
<i>Spörndly E.</i>	
Nutritive value and composition of vegetation selected by steers grazing semi-natural pastures every other year compared with every year .....	1010
<i>Spörndly E.</i>	
Improvement of oligotrophic pastures with animal and mechanical treatments .....	1013
<i>Staglianò N., Argenti G. and Sammarone L.</i>	
Effect of two different supplementary feed rations on pastures in an area of the Italian Alps .....	1016
<i>Staglianò N., Seppoloni I., Clementel F. and Argenti G.</i>	
Linking <i>Senecio aquaticus</i> occurrence to grassland management .....	1019
<i>Suter M. and Lüscher A.</i>	
<b>Index of Authors .....</b>	<b>1023</b>

# Opening Session

## Biodiversity and animal feed



# Semi-natural grasslands in Europe today

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## Abstract

Semi-natural grasslands have for a long time had low priority in European nature conservation. Since World War II there has been a rapid loss of semi-natural grasslands in Western Europe. In 2004 the EU began to make serious efforts to evaluate the areas of semi-natural grasslands within the EU, but the presented values are only rough estimates. In the Benelux countries, Denmark, northern France and Finland the situation is rather grim, while Sweden and the UK have a more favourable situation. In the Mediterranean countries, there are still considerable grassland areas remaining. Spain is the individual country within the EU with the greatest acreage of semi-natural grasslands. Since the fall of the Soviet Union, grazing pressure on the landscape has decreased radically, and this has benefited some species, e.g. steppe birds such as cranes and great bustards. International cooperation can lead, hopefully, to Russia taking better care of its semi-natural grasslands, as has already happened in e.g., Hungary and Estonia. Apart from Spain and Russia, Romania is probably the country that has the greatest well-managed semi-natural grasslands in Europe. The values of semi-natural grassland are summarized in terms of four categories. Hypotheses about the evolution of grassland species before the agricultural landscape developed are also discussed.

Keywords: semi-natural grassland, pasture, biological diversity, grazing cattle, management, recreational value, cultural heritage

## Introduction

### *Semi-natural grasslands not a European focus*

Semi-natural grasslands have not been an area with which conventional European nature conservation has particularly concerned itself. Forests, mountain areas and wetlands have been more in focus. In addition, many of the semi-natural grasslands of Europe have been regarded as the unhappy degeneration products of hypothetical mighty forests, the loss of which has been deeply regretted. In the tradition of the wilderness that has come to strongly affect European nature conservation during a large part of the 20th century, largely through the influence from the USA, grasslands even came to be regarded as areas on which these disappeared forests should be restored. Maccia, the shrub-rich grasslands in the Mediterranean area, came to represent worthless wasteland, created through mankind's thoughtless over-exploitation, e.g. by overgrazing, and nature conservation experts for a long time propagated for replanting of maccia areas.

### *Some early attention*

In those areas of Europe in which modernization of land use occurred earliest, e.g. England and Denmark, attention was drawn relatively early to certain types of indigenous semi-natural grassland that had begun to become rare through cultivation and replanting. Heather moors came to attention in this way. In north-west Germany for example, the Luneburger moors came to acquire a romantic character and have therefore been partly saved.



### *Rapid grassland loss*

Since World War II there has been a rapid loss of semi-natural grassland in Western Europe. Planting of forests and cultivation, i.e. fertilization and sowing of highly productive grass and legume leys, quickly led to the disappearance of large areas. Sweden, together with the United Kingdom and the Netherlands, was one of the countries that began in the 1980s to carry out a national inventory and also a conservation programme for their semi-natural grasslands. Despite great efforts, the area of semi-natural grasslands has decreased even in these countries, but the very negative trend has been broken and, at least in terms of area, the situation is almost stable, while some semi-natural grasslands in marginal areas of Sweden and the UK have ceased to be managed.

### *Grassland areas in the EU*

Only in the past ten years have semi-natural grasslands been given more obvious attention at a European level. It was as recently as 2004 that the EU, through EEA (the European Environmental Agency in Copenhagen), began to make serious efforts to evaluate the areas of semi-natural grassland within the EU. The values that the EEA presents are only estimates, as it fully acknowledges, and sometimes the span between the highest and lowest value is very large, as in the case of France where it is specified that between 3 and 27% of the entire agricultural area can be regarded as 'high nature-value farmland', which in reality consists of different types of grassland. Although the uncertainty is greatest in the case of France, it is also very great for other EU countries, and it is not only a question of difficulties in measuring areas, but also to a very high degree a problem of definition. For example, what level of fertilization is acceptable for an area of grassland to be counted as semi-natural grassland? When does a grazed arable fallow become a semi-natural grassland that is valuable from a nature conservation perspective? Is it always inevitable that such grasslands should be assessed on botanical merits? Can arable fallow with a high ornithological value, such as that in Hungary, be given a high rating? How should the grazing land around the treeline in mountain areas be considered?

### *Statistical variations*

An interesting observation when working with statistics and other material on European semi-natural grassland is that in many ways we have a better idea of the current situation in the new EU member countries than in the original 15. This is probably because those countries had pressure placed on them before EU entry to provide a good picture of the situation, in that they were expected to develop national applications of the EU environmental subsidy system.

## **The grassland situation in Europe**

What is the situation like in Europe today for semi-natural grasslands and similar areas?

### *Northern and north-west Europe*

In north-western areas such as the Benelux countries, Denmark, northern France and Finland the situation is rather grim, even though intensive work is being carried out on what remains. Very small areas of very valuable and well-managed semi-natural grasslands remain here. The exception is certain areas with coastal grasslands. Understanding of using grazing is even limited in nature reserve management. Recently published studies from southern Germany show that practically all nature reserves in that area with a grazing-adapted flora and fauna lack a grazing system. Mowing has been used as an alternative, with very variable success. In northern Europe, it is Sweden and the UK that have the most favourable situation. In terms of area the UK considerably exceeds Sweden, but there is a statistical anomaly which means that in the British Isles a large proportion of moorland and even partly fertilized grassland is

included in the figures. Norway, which is traditionally regarded as a country with large semi-natural grassland resources, has no current national inventory and the situation there, assessed on the basis of spot sampling, can be regarded as very serious. Large areas of traditional grazing land have disappeared in recent decades.

#### *The Mediterranean situation*

Further south, in the countries around the Mediterranean, there are still considerable areas of semi-natural grasslands remaining. However, the situation there has also deteriorated rapidly during the past twenty years and it is only very recently that the semi-natural grasslands in these areas have begun to receive some attention. Spain is the individual country within the EU with the greatest acreage of semi-natural grasslands. For example, in recent years increasing attention has been devoted to the dehesa grassland in Spain, which has its equivalent in the montado in Portugal. Dehesa, which is mainly found in south-western Spain, is a savannah-like grazing landscape with occasional wide-crowned cork oaks and holm oaks. The grassland is cultivated now and then, and cropped for a few years before being returned to grazing. The oaks produce acorns that are of benefit as pig feed but also for overwintering Nordic cranes. Dehesa grassland formed part of a transhumance system, i.e. a grazing system whereby the dehesa was used for sheep grazing in the winter, after which the sheep were moved to mountain grazing, e.g. in the Cantabrian Mountains. Similar transhumance systems persist on a greater or smaller scale in southern Africa, Italy and Greece. In northern Greece in particular, there are still extensive areas of semi-natural grassland that are used in summer, while in winter the grazing animals are moved to near the coast.

#### *The situation in the former Soviet Union*

Since the fall of the Soviet Union, agriculture in central and Eastern Europe has changed significantly. Different degrees of privatisation have occurred, but the entire agricultural sector has also generally decreased as a result of competition from the West, and from outworn and outdated production equipment and great uncertainties regarding land ownership. Grazing pressure on the landscape has thereby decreased radically and large areas have been left unmanaged. As mentioned above, comprehensive inventories have been carried out with aid, e.g. from the Netherlands, on the scope and status of the grasslands in Eastern Europe. It can be concluded that there are significant areas remaining in the East but very large areas are unmanaged. In Russia the fall of communism has brought about a decrease in the earlier unreasonably severe grazing exploitation, which has benefited some species, e.g. steppe birds such as cranes and great bustards. However, Russia has no clear official nature conservation ambition as regards its semi-natural grasslands, which is regrettable in view of the very large areas of interesting grasslands that exist in southern Russia in particular. International cooperation can hopefully lead to Russia taking better care of its semi-natural grasslands, as has already happened in e.g. Hungary and Estonia.

#### *High grassland-values in Romania*

Apart from Spain and Russia, Romania is probably the country with the greatest well-managed semi-natural grasslands in Europe. There is also a large variation as regards semi-natural grasslands in Romania, from dry steppes in the East through wet grazing lands along the banks of the Danube and in the Danube delta, to huge grasslands on the slopes of the Carpathian Mountains. Unfortunately, these large natural resources are partly the result of the poverty caused by the Ceausescu dictatorship. The country is now a member of the EU and it faces a great challenge in avoiding losing its large natural and cultural resources while at the same time implementing an essential modernization of agriculture. Environmental subsidies on a large scale will be required in Romania. However, people do not always sit and wait for

the authorities to react and provide them with good conditions. There are already examples in Romania of local initiatives to utilize the value contained in semi-natural grasslands in a positive way, as in the case of the Botiza village in Maramures in north-west Romania where ecotourism has begun to develop. The village is very much alive and has around 150-ha arable fields that partly move around in the meadows, which are dotted with trees and shrubs which are pruned in different ways. The total area of meadow, which is hand-mown by scythe, is about 3000 ha. That is a greater area of dry meadow than exists in all of Sweden.

There are still high values in Europe, but preservation efforts are needed. Europe is still a goldmine for those wanting to experience different types of semi-natural grasslands and meadows. However, great efforts are needed if future generations are to be able to enjoy these fantastic areas in Spain, Sweden, Russia and Romania.

## Discussion

### *Four categories of European grassland values*

Why are the semi-natural grasslands of Europe so valuable? Their value can be summarized in terms of four categories.

(1) On semi-natural grassland, meat and dairy products can be produced in a way that is highly acceptable from an animal ethics perspective. From a general environmental perspective too, production occurs in a way that has little or no negative impact on the surrounding landscape. On the contrary, the semi-natural grasslands contribute to the highest degree to the general 'high standard' of many landscapes. Meat and dairy products from semi-natural grassland can therefore be an alternative for consumers, and can be regarded as superior to vegetarianism in terms of animal ethics and environmental considerations.

(2) The biological diversity in European semi-natural grasslands is very high. Very large proportions of Europe's most threatened bird species, vascular plants and insects live in these grasslands. Their future existence is threatened if we are unable to manage and preserve these areas. For threatened species in e.g. a number of natural forests, the problem is actually easier to solve. Here nature reserves can be established and forest operations can be more or less stopped, but semi-natural grasslands require management, and management requires active farmers who can fully or partly make a living from these grasslands. The structure of future European countryside policy has a decisive impact on the future fate of semi-natural grasslands and thus for the threatened species living therein. This is particularly true for eastern areas of Europe.

(3) Semi-natural grasslands can in many cases have a very high recreational value. A number of studies have shown that the 'pastoral landscape' is high on the list for many people as regards their preferences for, e.g. recreational activities.

(4) From a cultural heritage perspective semi-natural grasslands play an important role in many European countries. Often little thought is given to the fact that much of the regional identity presented in, for example, a tourism context, is founded on semi-natural grasslands and the traditional use of such lands. One only has to think of the heather moors of Scotland, the dehesa of southern Spain, the mountain pastures of the Alps and the shore meadows of south-western Denmark.

However, the cultural heritage perspective extends beyond this; and this brings us to a very exciting and partly unresearched area of science. This problem area can be summarized thus: *Where were all the animals, plants and fungi that live in the semi-natural grasslands of Europe today, and that would die out if we were to cease taking care of these grasslands, where were they living before Europe had farmers and herders?*

The answer is naturally complex and includes at least the following components. Certain species live in other types of habitat, e.g. above the treeline in alpine areas, on sea shores or in natural indigenous areas. Others have migrated in from semi-desert areas in African and Asia. Some even appear to have been created through evolution during the relatively short time period that farming has existed in Europe, but there are few such species and they are often a case of varieties and landraces. Finally, we come to the part of the answer that is the most debatable but also the most exciting: there were wild grazing animals that created a pastoral landscape in Europe before farmers with their domesticated animals brought about a similar landscape. The wild ox, ancestor of the modern cow, which is now extinct, was probably the most important species in this regard.

#### *A challenged hypothesis*

There is no scientific agreement concerning the above hypothesis and it has been challenged on a range of issues. However, it is clear that many of the species that are found today on the semi-natural grasslands of Europe have their evolutionary background in natural ecosystems that were very strongly influenced by large grazing mammals. The 'baton' must have passed from wild grazing animals to tame, and the grazing-dependent species have in many cases succeeded in leaping over from a system without human influence to a system created and controlled by humans. Exactly how this has occurred is still very unclear, e.g. this 'baton change' may not necessarily have taken place in north-western Europe but in other areas, after which the entire 'package' of grazing animals and grazing-dependent organisms accompanied humans to north-western Europe. The grassland areas in today's Hungary and Romania may have played an important role in this 'baton change'.

#### *Grasslands with both biological and cultural values*

Whatever the case, I believe that the species of semi-natural grasslands are very well worth protecting, since although they live in a system strongly influenced by humans, it is a system with very old cultural traditions. These species are no less valuable than those living in the wild. In fact, they have both a purely biological value and a cultural history value. In addition, they perhaps also represent a type of nature that we humans have eliminated through eradicating the wild ox, the wild horse, the giant elk and a range of other large grazing animals. The animals, plants and fungi of semi-natural grasslands have been able to survive thanks to humans having replaced the wild grazing animals they eliminated with domesticated versions.

#### **Concluding remarks**

Much research remains to be done into the fascinating history of semi-natural grasslands but, whatever the answers, they are also very much an issue of our own history. I believe it is time for the cultural history of these areas, their domesticated grazing animals and their wild animals, plants and fungi to be given greater attention. Some attention should also be devoted to the trees and shrubs of these areas, which have always played a very important and complex role. There are sometimes obvious signs that the value of trees and shrubs has not been understood by EU countryside experts, e.g. in the structure of the regulations for environmental subsidies that are currently essential for the conservation of Europe's semi-natural grasslands.

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# Challenges for grasslands, grassland-based systems and their production potential in Europe

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## Abstract

The importance and the diversity of grasslands and grassland-based systems are illustrated. Their multiple functions and values are highlighted. The main trends of the evolution of the systems in the last half-century are summarized including the effect of the Common Agricultural Policy of the European Union. Grasslands are compared with annual forage crops for forage production potential and impact on the environment. New challenges are analysed especially in relation to agro-fuel production. The potential of grasslands for fuel production is discussed as well as the possible effect of first generation agro-fuels on the grassland area. The fractionation of grassland biomass into several products like fibres, lactic acid and amino acids is described in relation to possible industrial uses. Policy recommendations are proposed.

Keywords: feeding quality, multi-functionality, agro-fuel, biomass, green bio-refinery, agricultural policy

## Introduction

Grassland is by far the first land use in the agricultural area (AA) in Europe. Grasslands and rangelands cover 56 million ha (33% AA) (EU27, in 2005). This area includes about 17.5 million ha of rangelands (10% AA), mainly in mountain areas (EUROSTAT, 2008). This average importance of grasslands hides big differences between Member States of the EU. In West Europe, the proportion of grasslands in the AA is usually higher, as in the UK (62%) and Ireland (73%), while in Eastern Europe the proportion is usually lower, such as Poland (21%) and Romania (33%). That reflects the differences of ecological conditions, production systems, living standards, history and policies between countries.

Diversity is a keyword for grasslands and grassland-based systems in Europe. It is induced by the variability of the ecological conditions (soil, climate), species composition, use, management system and performances.

Grassland plant communities are classified in the European Union into seven main habitats according to EUNIS (2006): dry grasslands, mesic grasslands, seasonally wet and wet grasslands, alpine and subalpine grasslands, woodland fringes and clearings and tall forb stands, inland salt steppes, and sparsely wooded grasslands. Grassland swards can be species-rich or species-poor, dominated by grasses, by legumes, or by a balanced mixture of grasses and legumes. In some swards, usually species-rich, these two plant families can even have a minor importance compared with other species types.

Grasslands may be permanent or temporary (regularly sown). They can be only grazed, only cut or mixed used for grazing and cutting. The use of inputs per ha (fertilization, amendments, herbicide use, re-sowing or over-sowing, irrigation) in grassland can be highly variable, much more than for arable land. The large variation in input use, management, vegetation types and ecological conditions induces large differences in dry matter production, forage quality, stocking rate and animal production. Low-production swards can only produce annually about 2-3 t DM ha<sup>-1</sup>; they can be grazed by sheep for instance. High-production swards can yield as much as 10-12 t DM ha<sup>-1</sup> for grazed swards and 15-20 t DM ha<sup>-1</sup> for cut swards under good

production conditions; they are usually used for dairy cows. A poor-quality, low-digestibility hay has high fibre (CF about 35-40% DM) and low protein contents (CP about 6.25% DM) while a highly digestible young grass is fibre-poor (CF about 15-20% DM) and protein-rich (CP about 18-22% DM). Some mountain swards for instance can only feed 2-3 ewes (0.17-0.25 Livestock Units (LU)) per ha during the summer months while a productive lowland sward can be stocked with 5-6 dairy cows (or LU) in spring and 3-4 cows (or LU) in summer. In North-West European countries, where grassland yields are the highest without irrigation in Europe (Peeters and Kopec, 1996), the annual milk yield is estimated at 9,000 to 13,000 kg ha<sup>-1</sup> of forage crops (Kristensen *et al.*, 2005). Marginal grasslands can just cover the maintenance needs of low-requirement animals. This range of values has no other purpose than illustrating the wide variability of grassland production potential in Europe.

Grassland-based systems are also highly diverse according to the main animal production. Dairy systems are the most intensive ones while meat-sheep systems are usually the most extensive ones. In some systems, animals are kept indoors all year long while sheep and heifers for instance can stay outdoors at all seasons. Pflimlin *et al.* (2005) identified 7 forage and livestock regions in the EU15: 1. Northern regions (Scandinavia); 2. Wet mountain regions (Alps, Vosges, Jura, Massif Central, Pyrenees, Cantabrian mountains); 3. Mediterranean regions in fertile plains and valleys as well as in dry mountain rangelands; 4. Grassland regions (permanent grassland dominant in the AA) (examples: Ireland, Wales, Scotland, South and North West of England, Belgian Ardennes, North East of France); 5. Grassland and maize regions (e.g. North West of Germany, East of the Netherlands, 'Bocage' of Normandy); 6. Forage crops regions (temporary grasslands + maize) (e.g. Brittany, Belgian Flanders, South East of the Netherlands, West of Galicia); 7. Arable land and livestock regions (e.g. margins of the Parisian Basin, North of the Pô plain, part of Bavaria, central part of England). The majority of livestock holdings (60%) are located in Less Favoured Areas (LFA) (Mountain, Mediterranean and Grassland regions). Livestock systems are using at least 42% of the EU15 agricultural land but probably much more (Vidal, 2001).

Grasslands are characterized by multiple functions and values. They are providing forage for grazing and browsing animals, both domestic and wild. Compared with high-density coniferous tree plantations, they have a positive influence on the recharge of water tables. Compared with annual crops, they have a protection effect for water quality and a good potential of carbon sequestration in the soil. They protect the soil from wind and water erosion, and enhance soil fertility. They are the support of an important biodiversity; some extensive grassland types have a very high nature value. They are supporting rural economies and are a source of livelihood for local communities. Grassland landscapes are aesthetically pleasing, provide recreational opportunities, open space and improve the quality of life of the whole society.

An analysis of the outlook of grasslands and grassland-based systems must take this diversity and this multi-functionality into account. Changes in socio-economic factors have not the same effect on each of these systems. This paper focuses on the factors that can affect grassland area, grassland-based systems and their production potential in the future. It does not tackle specifically the environmental aspects. It analyses the possible future uses of grasslands in the context of energy and primary raw material production. Agro-fuel, bulk chemicals, human food, animal feed and fibre production could be developed from grassland forages in the future in addition to the traditional use of ruminant feed.

### **Main trends in the last half-century**

Two major trends characterized grassland systems in the last 50 years, i.e. roughly since the beginning of the Common Agricultural Policy (CAP) of the EU in the early 1960s:

intensification and land abandonment. In the lowlands, nitrogen fertilization in grassland started to be used at a large extent since the 1960s. Stocking rate, frequency of cutting for conservation, fertilizer use, drainage, irrigation, re-sowing and over-sowing with improved varieties, weed control with herbicides became increasingly important. The number of plant species (and biodiversity in general) fell dramatically in grassland swards while forage yields increased and feeding quality improved. At the same time, farm and farmer numbers were reduced and farm size increased. That modified the traditional landscape by an enlargement of plot size and, as a direct consequence, a decrease in field margins and edges. Between 1975 and 1990 (EU15), the grassland area was significantly reduced in favour to the production of fodder maize and cash crops. After 1989 and the fall of communist regimes, many agricultural areas and especially grassland areas were abandoned in countries in transition. It is estimated that at least 30% of grassland areas were abandoned in countries like Bulgaria and Romania. Actually, in some regions, including in mountain areas, the *quasi* totality of grasslands were abandoned. That can be illustrated by the dramatic decrease of ruminant numbers. In Bulgaria for instance, LU (cattle, sheep, goats) in 2005 were 34% of its value in 1985 (NSI Sofia, 2005). The same calculation for Slovakia between 1990 and 2004 (cattle, sheep) gives a similar value of 35% (NSI Bratislava, 2004). Even in the EU15 countries, marginal grasslands tended to be abandoned, especially in mountain areas. Intensification and abandonment are the two sides of the same coin. When yields are increasing, less acreage is necessary for achieving the total production and the production costs are increasingly important in marginal regions compared with favourable, intensively used regions.

The specialisation of productions resulted in the progressive disappearance of mixed farming. Some regions specialised in arable crops while other regions specialised in animal husbandry. The importance of temporary grasslands and especially *Medicago sativa* declined everywhere, even in grassland regions. In the 1950s and the 1960s, this trend was reinforced by the decline of agricultural manpower that made traditional haymaking very difficult. The use and the proportion of legume species (especially *Trifolium repens*) in swards were also reduced by a widespread use of nitrogen fertilizers. Animal breeds were specialised for milk or meat production, while dual-purpose breed populations were reduced. Dairy systems were concentrated in the lowlands (74% EU dairy cows) especially in Atlantic climates (regular rainfall and mild temperatures are ideal for grass growth), and also at medium altitude in mountain areas (11% EU dairy cows) (Pflimlin *et al.*, 2005). Beef meat systems occupied more marginal soils and climates. As a consequence of specialisation, animal performances increased. In dairy systems, from an average of 4,500 l per cow and per year in the 1970s, the annual production increased to 7,500 l/cow while some herds or some cows are reaching now an annual production of 10,000 to 12,000 l/cow. This regular increase of dairy performances of about 1% per year was possible thanks to an international effort of breeding of a restricted number of dairy breeds among which the Holstein breed is undoubtedly largely dominant. That led to a decrease of the populations of many less performing breeds. Yield increases induced also changes in animal feeding. More concentrates were used at the expense of green forages. The implementation of milk quotas in 1984 slowed down this trend because the control of the volume of production required a decrease of production costs. This was achieved by a better utilisation of green forage that are less expensive than concentrates and by an even higher production per cow. Indeed, this made it possible to decrease the share of maintenance feeding needs and to increase the share of production needs. More milk had to be produced per cow on the basis of grass and maize. However, the CAP reforms of 1992 and 2000, induced a significant decrease of cereal price (about 50%) and that encouraged again dairy farmers to use this product in animal feeding. Moreover, farmers tended to use more maize silage at the expense of grass grazing and grass silage when dairy cow production is above a certain threshold (roughly above 6,000 l/cow). They did not trust grass quality and



grass intake potential of their high-yielding cows especially by rainy weather and unfavourable temperatures. They tend thus to keep partially cows indoors or to complement systematically grass grazing with maize silage. That led to a decrease of grassland proportion in the AA. This trend was very strong in the 'Forage crop and the Grassland and maize regions'. In the 'Grassland region', the strategy of farmers was a bit different; they used less maize but increased concentrate use. They also tried to better reduce production costs by a better use of grazing and grass silage. In the 'Wet mountain region', farmers have smaller herds, use more green forages including grass, and they can increase their income by selling high-quality cheeses promoted by a 'Protected Designation of Origin' (PDO) system at a high price. In beef production systems, a small number of good conformation breeds emerged too. Local traditional breeds tended to be progressively crossed with three dominant breeds: Charolais, Limousin and Belgian Blue. Grazing remained the basis of suckling cow systems and animals were fed in winter mainly with hay and haylage. Concentrates and maize silage were restricted mainly to bull fattening. Ox fattening disappeared almost in favour of young bulls. In Mediterranean regions where most sheep and goats are located, the number of grazing animals decreased leading to a large abandonment of dry rangelands. All these system changes had impacts on landscape and wildlife by reducing diversity and complexity. In the 'Forage crop region' and in some intensive parts of the 'Grassland and maize region', farmers had to face criticisms for their negative effect on the quality of ground- and surface-waters. Measures had to be taken to decrease nitrate and phosphate pollution.

The CAP expenditures are made of two main areas or 'pillars'. The first pillar provides income support through direct payments to farmers. It also includes, to a lesser extent, market support measures. The second pillar is devoted to rural development policies. These policies are aimed at providing assistance to difficult farming areas, encouraging environmental services, and promoting food quality, higher standards and animal welfare (EUROPA, 2004; CEC, 2007). Some of the measures of the second pillar were designed for correcting perverse effects of the first pillar. Rural development expenditures were increased after the reform of 2000. In 2006, they represented € 7.7 billion annually and 15% of the total CAP expenditures (€ 4.4 billion and 10% in 2002)(EUROPA, 2004). LFA payments contribute significantly to livestock farmers' income since more than one-half of them are operating in these areas and the amounts of payment are not negligible though much lower than those from the first pillar (Pflimlin *et al.*, 2005; Roeder *et al.*, 2007). While the annual payments from the first pillar sum up to roughly € 300 ha<sup>-1</sup> on average in the EU15, the LFA payments range from less than 25 to more than € 50 ha<sup>-1</sup> according to Member States (Table 1). These payments are often linked with specified stocking rates. Agri-environmental measures (AEM) were designed to encourage farmers to protect and enhance the environment on their farm. Farmers receive a payment in return for a service. Their commitment of improving the environment is only rewarded if it goes beyond the application of usual 'Good Agricultural and Environmental Practices' (GAEP). Some measures tried to promote grassland areas and limit the increase in maize and cash crop areas but they were not able to reverse the general trend. They probably decreased the reduction rate of biodiversity in grassland and the simplification of landscapes. Agri-environmental payments remained relatively modest. In 2002, the EU15 spent 2 billion euros for AEM implementation, i.e. about 4.6% of the total amount of CAP funds. In 2000-2003, 16.3 euros were spent, on average, per ha AA of the EU for AEM. It reached 89 euros per ha AA in Austria. An average of 89 euros were received by EU farmers per ha under AEM contract. In 2002, the share of agricultural land enrolled in AEM in the EU15 reached about 25% of the AA but it varies from less than 5% in the Netherlands and Greece to more than 80% in Austria, Sweden, Finland and Luxemburg (EEA, 2006). AEM included the support of the conversion to Organic Farming (OF) and in some Member States to the maintenance of OF. OF is characterised by a mixed farming (crop and animal husbandry) and

is based on a large use of grassland and forage legumes. It has developed rapidly since the implementation of the AEM in 1992, with more than 5.8 million ha, 3.4% of the AA and almost 140,000 organic farms in 2004 (EEA, 2007). Except in some regions and Member States, it remains marginal and did not change the main evolutionary trends of the EU agriculture. The new Rural Development policy 2007-2013 focuses on three areas: improving competitiveness for farming and forestry; environment and countryside; improving quality of life and diversification of the rural economy. A fourth axis called 'Leader axis' based on experience with the Leader Community Initiatives introduces possibilities of direct support to locally based bottom-up approaches to rural development (EUROPA, 2007).

Table 1. Importance of LFA annual payments in the EU15 (Roeder *et al.*, 2007).

€/ha	% of AA within LFA		
	< 20	20-60	> 60
< 25	BE, DK, NL	IT	ES, PT
25-50		FR, SE, UK, DE	
> 50		IE	AT, GR, FI, LU

Sources: EEA (2004, 2005); EUROSTAT (2005).

The CAP reform of 2003 introduced four new principles into the previous systems: decoupling, cross-compliance, modulation and partial re-nationalisation. With regard to grasslands, these modifications had some important consequences. Decoupling induced that premiums were not linked with crop or animal types but to the eligible area. That suppressed the 'maize premium' that encouraged farmers to use this forage crop at the expense of grasslands. The use of grasslands was also not anymore indirectly supported through animal premiums but directly through area payments (the system was though applied with a certain amount of flexibility among Member States according to the re-nationalisation principle). This measure reduced the distortions that were unfavourable to grasslands. Cross-compliance required that farmers must comply with a set of GAEP, including the obligation to maintain the proportion of permanent grassland in the AA, in order to maintain their land in 'Good Agricultural and Environmental Conditions' (GAEC). Farmers have also to respect the Habitat, Bird and Nitrate Directives. The Nitrate Directive is mandatory for farmers and had a significant influence on farm structures and practices of intensive livestock systems by regulating the stocking rate and the management of nitrogen. The modulation principle intended to transfer a part of the first pillar budget to the second pillar. Partial re-nationalisation concerns notably the way Members States can define the GAEC and the eligible area. It has also an impact on the implementation of the modulation principle.

The successive EU CAP reforms led to modernisation of the sector, farm size increase, a dramatic farmer-population decrease, specialisation of productions, intensification of grassland and animal husbandry, increase in the volume of production, increase of grassland and animal yields, reduction in legume use, reduction in grassland area and its proportion in the AA, reduction in the diversity of landscape, grassland species and communities, domestic animal breeds and local products. The implementation of milk quotas reduced the number of dairy cows which induced stocking rate decrease in some cases or the development of suckling cows systems in complement to dairy systems in other cases.

In addition to agricultural policies, a social factor had an influence on grassland systems: the constant reduction in beef and sheep meat consumption of European citizens in favour of pig and poultry meat. If less ruminant meat is consumed and the grassland area does not change, an extensification of grassland management is possible, but it is more likely that a higher demand for monogastric meat will induce replacement of a part of the grassland area by crops.

## Grassland *versus* other crops for forage potential

In order to understand and to forecast the future potential role of grasslands in the European farming systems, it is necessary to compare grasslands with its direct alternatives for forage production. Table 2 summarizes typical yields of the main forage crops, forage maize and fodder beet, compared to intensive cutting temporary grasslands and grazed permanent grasslands in the conditions of North-West Europe. Dry matter yields of fodder beet are usually higher than those of maize. Yields of temporary grassland under cutting are very high too, but on average a bit lower than maize. Grazed permanent grasslands have lower yields notably because of frequent defoliations and trampling. Table 3 compares typical feeding values and intake characteristics of these forage crops. The energy content is the highest for fodder beet followed by maize and then by grass. Grass has higher protein and mineral contents than maize and fodder beet. The voluntary intake of fodder beet is excellent but it must be limited for controlling acidosis of the rumen. Intake of maize silage is very good, usually better than that of grass silage, but similar to that of grazed grass.

Table 2. Comparison of typical yields of the main forage crops in North-West Europe (Deprez *et al.*, 2007).

	Forage maize	Fodder beet	Cutting temporary grassland	Grazed permanent grassland
t FM/ha	50	120	50	57
%DM	30	13-19	20-30	15-20
t DM/ha	13-18	14-21	12-16	8-12

FM = fresh matter; DM = dry matter.

Table 3. Comparison of typical feeding value and intake characteristics of the main forage crops in North-West Europe (Deprez *et al.*, 2007).

	Forage maize	Fodder beet	Cutting temporary grassland	Grazed permanent grassland
Energy/kg DM				
UFL	0.90	1.15	0.90	0.80-0.98
UFV	0.80	1.16	0.84	0.83-0.94
Protein (g/kg DM)				
CP	70-85	50-100	120-170	120-240
DP	45-55	65	70-100	80-120
PDIA	20	10	25	27
PDIN	50	60	85	75-115
PDIE	65	85	75	80-110
Minerals (g/kg DM)				
P	1.6-2.8	1.5-2.0	2.8-4.0	3.0-4.8
K	12-16	22	22-30	25-40
Na	0.05-1.0	1.0	3.0-4.0	1.0-2.0
Ca	2.5-4.4	2.0-2.5	3.0-6.0	5.5-9.4
Mg	1.5-1.8	1.5	1.5-3.0	1.3-2.5
Digestibility (%)	68-73	90-94	68-72	70-80
Intake (g DM/kg LW)	23-30	Excellent but must be limited (acetonemia)	16-20	20-33

UFL = Unité Fourragère Lait (Fodder Unit milk) = Net Energy content of 1 kg of barley for milk (1700 kcal NEI); UFV = Unité Fourragère Viande (Fodder Unit Meat) = Net Energy content of 1 kg of barley for meat (1,820 kcal NEg); CP = Crude protein; DP = Digestible Protein; PDIA = Protéines Alimentaires Digestibles dans l'Intestin grêle = Feed Protein ruminally undegraded and truly Digested in the small Intestine; PDIN = Protéines Digestibles dans l'Intestin grêle avec azote limitant = Protein truly Digested and absorbed in the small Intestine when a degradable N deficient diet is fed; PDIE = Protéines Digestibles dans l'Intestin grêle avec énergie limitante = Protein truly Digested and absorbed in the small Intestine when a ruminal fermentable Energy deficient diet is fed.

Conserved and grazed grasses are complete feeds, rich in energy, protein and minerals. Grazed grass is even too rich in protein and in K. It must be complemented by energy-rich and protein-poor feeds. Maize silage is energy-rich, highly digestible and has very good intake characteristics but it is deficient in protein. This table demonstrates the complementarities between grazed grass and maize silage on the one hand, and grass and maize silages on the other. Table 4 evaluates the ease of the main forage crops for cropping, conservation and animal feeding. Maize silage is easy to crop, to harvest and to conserve. Grass silage is easy to crop, but more difficult to harvest because it requires several cuts per year. It is also poorer in soluble carbohydrates than maize and is thus a bit less easy to conserve. The distribution is similar. All technical aspects are difficult to control for fodder beet.

Table 4. Evaluation of the easiness of the main forage crops for cropping, conservation and animal feeding (Deprez *et al.*, 2007).

	Forage maize	Fodder beet	Cutting temporary grassland
Cropping	++	-	++
Harvest	++	-	+
Conservation	++	-	+
Distribution	++	-	++

++: very easy; +: relatively easy; -: difficult.

Table 5. Parameters of forage crop profitability (for Belgium in 2005) (Deprez *et al.*, 2007).

	Forage maize	Fodder beet	Cutting temporary grassland	Grazed permanent grassland	Species-rich hay meadow
Farm rent (€/ha)	150	150	150	135	135
Installation costs (€/ha)					
Seed bed preparation	125	125	120	-	-
Sowing	50	50	100	-	-
Seeds	165	125	75	-	-
Maintenance	-	-	-	150	75
Total	340	300	295	150	75
Inputs (€/ha)					
Fertilizers	270	230	275	90	-
Herbicides	100	210	30	-	-
Insecticides		20			
Total	370	460	305	90	0
Harvest costs (€/ha)					
Cutting-ensiling	265	340	650	-	-
Plastics + silo	150	75	130	-	-
Total	415	415	780	0	135
Annual yields					
t DM/ha	17	18	14	10	4
UFL/t DM	940	1100	830	900	750
kg CP/t DM	70	75	130	140	100
Cost price without premium (€)					
per ha	1275	1325	1309	375	345
per 100 kg DM	7.50	7.36	9.35	3.75	8.63
per UFL	0.08	0.07	0.11	0.04	0.12
per kg CP	1.07	0.98	0.72	0.27	0.86

Table 5 compares the profitability of the same forage crops and additionally of hay from species-rich meadows. It appears that annual crops and cutting grasslands have similar costs per ha (slightly higher for grass silage) and much higher than grazed grasslands and hay meadows. Temporary grasslands and hay meadows have the highest costs per kg DM and per

energy content and grazed grasslands the lowest ones. All kinds of grasslands and especially grazed grasslands have the lowest costs per kg of crude protein.

### Grassland *versus* other forage crops for the impact on the environment

Tables 6 and 7 summarize the effect of different forage crops on some important parameters of the environment: pollutions by nitrate, phosphate and pesticides, as well as biodiversity conservation. Table 6 shows that all types of intensive grasslands have lower risk of pollutions than maize and fodder beet. Nitrate leaching can though occur after the destruction of temporary grasslands if basic rules are not respected. The most intensive grazed grasslands can also contribute to nitrate leaching especially if fertilized after the end of August and heavily stocked. Phosphate and nitrate pollutions of surface water can occur in grasslands with the access of animals to rivers and with manure applications on slopes and in rainy periods or in winter. Annual fodder crops present higher risks of pollution (except fodder beet for nitrate leaching) including for pesticides that are not used, or little used, in grasslands. Table 7 illustrates that all types of biodiversity can better develop in grasslands compared with annual fodder crops that can be compared to 'deserts' for biodiversity. Species-rich grasslands have of course the highest nature value but they are much less productive and have lower digestibility and intake values.

Table 6. Pollution risk at plot level for the main forage crops (Deprez *et al.*, 2007).

	Forage maize	Fodder beet	Cut temporary grassland	Grazed permanent grassland
Nitrate	---	+	+ to -	+ to --
Phosphate	---	--	+	+ to --
Herbicide	---	-	+	+
Insecticide	-	--	+	+

---: very high risk; --: medium to high risk; -: medium to low risk; +: low or no risk.

Table 7. Synthesis of the impact of several forage crops on biodiversity (Deprez *et al.*, 2007).

	Forage maize	Fodder beet	Cut temporary grassland	Grazed permanent grassland	Species-rich hay meadow
Agricultural biodiversity	0	0	++	+	0 to +
Functional biodiversity					
Flora	0	0	+	++	+++
Soil activity	0 to +	0 to +	++	+++	+++
Insects	- to 0	- to 0	0 to +	++	+++
Patrimonial biodiversity					
Flora	--	--	- to 0	++	+++
Insects	-- to -	-- to -	0 to +	+ to ++	+++
Birds	- to 0	- to 0	0 to +	+ to ++	+++
Mammals	- to 0	- to 0	+	++	++

--: very negative impact; -: negative impact; 0: neutral effect; +: positive impact; ++: very positive impact; +++: extremely positive impact.

### SWOT analysis of grasslands compared to maize for forage production

A SWOT analysis is based on the study of Strengths, Weaknesses, Opportunities and Threats. Strengths of grasslands compared to maize are good DM yields of intensive swards, excellent grazing quality, high protein and mineral contents, permanent soil cover, low production costs, no or little pesticide use, possibility of variable use of fertilizers, usually acceptable levels of nitrate and phosphate pollutions, soil and water protection, positive or very positive effects on biodiversity. Grasslands (grass-legume mixtures) are one of the pillars of organic farming. Dairy and possibly meat products from grassland-based systems can be more tasty



and sometimes even more healthy than products from maize-based systems, for instance for PDO products. Extensive grazing systems can indeed be associated with high-quality products protected by labels. Grassland systems can produce high quality landscapes that can be associated with (agri-)tourism activities.

Weaknesses of grasslands are related to the fact that they must be harvested in several cuts, grass conservation relies on weather conditions, feeding quality and intake (grazing and silage) can be reduced in case of unfavourable weather conditions, costs for silage making are relatively higher, nitrogen content can be too high in some feeding systems especially during grazing, nitrate pollution risk after sward destruction (especially after ploughing) of temporary grasslands can be important, intensive cutting systems can be very detrimental to wildlife, intensive grazing systems can produce nitrate and phosphate pollutions as well as drastic wildlife reduction.

The disappearance in 2003 of the 'maize premium' of the former CAP system is an opportunity for grasslands. The protection of the grassland area is integrated in the present CAP cross-compliance system (GAEP). There is a social demand and a political willingness to protect the environment, landscape and biodiversity. There could be an increasing demand for meat and dairy products on the world market in the future. An increase of fossil energy and input prices can increase the attractiveness of grass-legume mixtures. The present increase of cereal price can induce higher and better use of green forages including grasslands for animal feeding.

A reduction of agricultural supports could be a threat in the future as well as a stagnation or a reduction of the importance of the rural development policy (second pillar) compared with the first pillar of the CAP. A significant proportion of the grassland area could be destroyed for agro-fuel production, especially with first-generation agro-fuels and for bio-gas production. An increase of grain and other crop prices could encourage farmers to plough grasslands for crop production. A decrease of the consumer's purchasing power could reduce beef and sheep meat as well as high-quality dairy product consumption.

The above data and information show the important potential of grasslands compared with other forage crops. This potential is still underestimated, including by agronomists and farmers. It is thus essential to advertise it and to promote the use of grasslands much more than it is done today.

## **New challenges**

After a period of over-production and control of the productions in Europe by quotas, set-aside and decoupling, new demands for cereals and other agricultural products are appearing on the World market because of the fast increase of the World human population and the emergence of new economic powers including China. The increasing price of fossil energy and environmental concerns about climate changes are also inducing crop-based agro-fuel production and demand. Both factors are leading to the increase of prices of agricultural products. Higher global demand for food has not yet had an impact on dairy and ruminant meat products in the EU but agro-fuel production has already induced a pressure on the European grassland area.

There are two major types of agro-fuels. Monocultures cropped on fertile soils can produce ethanol from starch or sugar of corn, wheat, barley, sugarcane and sugar beat, or bio-diesel from oil extraction from oilseed rape, oilpalm and soybeans mainly. Ligno-cellulosic agro-fuels can be produced from wastes such as straw, corn stove, or from annual and perennial crops like maize silage, C4 grasses (*Miscanthus* spp. in Europe), *Populus* spp., *Salix* spp. as well as from temporary (high yielding grasses and *Medicago sativa*) and permanent grasslands. The first agro-fuels belong to the so-called 'first generation' while the ligno-cellulosic ones are recognised as 'second generation'. First generation agro-fuels compete with

food production on good arable land, increase pollution from fertilizers and pesticides and threaten biodiversity by inducing higher intensification on the whole agricultural surface and by converting high nature value land into intensive agro-fuel productions. Some second generation agro-fuels like maize silage and, to a lesser extent, grass silage, mixed with slurry for methane production have the same effect. Other second generation agro-fuels and grasslands can be produced on marginal agricultural lands, are less competitive with food production, do not require (high amounts of) fertilizers and pesticides, and should have a better impact on biodiversity. Other agricultural wastes that have low ligno-cellulose contents like slurry, mixed with maize and/or grass silage, can be used for methane production.

When considering their full life cycle, there are increasing evidences that first generation agro-fuels (bio-ethanol and bio-diesel) in Europe are carbon sources. They release more greenhouse gases (GHG) during cropping, transportation and processing than the amount of CO<sub>2</sub> equivalent they fix. Effective agro-fuels should be carbon negative.

With respect to energy production, grassland biomass can be transformed into energy by several processes. Biological processes can be used for producing biogas (methane) by anaerobic digestion. Biomass can produce energy by direct combustion, notably in mixture with coal. It can also be transformed by gasification and other subsequent chemical reactions. Gasification relies on chemical processes at elevated temperatures (> 700°C) and includes pyrolysis. In a first reaction step, the ligno-cellulosic material is partially converted to CO and H<sub>2</sub> (syngas). In a second step, the syngas can be transformed into methanol, ethanol, mixed alcohols, hydrogen, Fischer-Tropsch diesel, or combusted for heat or electricity. The Fischer-Tropsch (FT) process is a catalysed reaction in which carbon monoxide and hydrogen are converted into liquid hydrocarbons of various forms. Combination of biomass gasification (BG) and Fischer-Tropsch (FT) synthesis is considered a very promising route to produce a synthetic fuel that can be used in diesel engines. All these transportation fuels are also referred to as 'biomass-to-liquid' (BTL) fuels.

Not all grasslands types have the same aptitude for bio-methane production. Methane yields are the result of two parameters (i) the specific methane production rate (m<sup>3</sup>/kg ODM) and (ii) the biomass yield (kg ODM/ha) (Taube *et al.*, 2007). It seems that grass species and cultivars differ little with respect to methane production rates. The development stage seems to have more influence; methane production rates seem to decrease with maturity and cutting number ranking. That has still to be investigated: the relation between digestibility and methane productivity seems not to be very close. Increasing crude fibre contents can increase methane production! Crude protein content has a negative effect which is a weakness of lucerne in this context (Eder, 2006; Gröblichhoff *et al.*, 2006). Typical methane annual yields of intensive grasslands would be of about 5,000 m<sup>3</sup>/ha, methane yields being much affected by biomass production. Maize and whole crop small grain cereals have comparable methane production rates to grasses but usually higher biomass yields. Typical methane yields of maize for instance range between 4,000 and 10,000 m<sup>3</sup>/ha. As a result, biogas farmers tend to use more maize than grasses and to convert permanent grassland to maize whenever possible. In this context, the use of grass silage for bio-methane production will be mainly restricted to swards that cannot be ploughed and in marginal environments in general.

Combustion of biomass from grassland is less favourable compared to other crops or residues like straw because of higher nitrogen, sulphur, chlorine and potassium contents (Taube *et al.*, 2007). Spring harvest of standing hay could reduce these contents but yield is then very much reduced (Hadders and Olsson, 1997). C4 grasses are better than C3 grass species because of lower ash contents. C4 grass cropping (*Miscanthus*) could be effective in this context.

Grassland biomass can be used as many other organic products for producing BTL fuels. However, by comparing biomass resources (straw, hay from surplus grasslands, wood residues from forestry and corn silage) in Baden-Württemberg (Germany), Leible *et al.*



(2005) concluded that the use of hay from surplus grasslands and corn silage is definitely less economic than the use of wood and straw. Fresh or conserved grass could though be used with an economic support because of the multiple functions that grasslands and grassland landscapes can provide to the society. A significant research effort is still needed to better define the possible use of grassland biomass by gasification and Fischer-Tropsch synthesis.

In the USA, Tilman *et al.* (2006) have shown that agro-fuels derived from species-rich grasslands (SRG)(including several C4 grasses) growing on degraded soils can produce more energy, greater GHG reduction and less pollution per hectare than corn grain ethanol or soybean bio-diesel. They calculated that SRG biomass burned with coal in electric generation facilities would produce a net gain of about 18.1 GJ ha<sup>-1</sup>, when converted in cellulosic ethanol and electricity about 17.8 GJ ha<sup>-1</sup>, via gasification and Fischer-Tropsch synthesis about 28.4 GJ ha<sup>-1</sup>. According to them, net energy gains from corn and soybeans from fertile arable land are about 18.8 and 14.4 GJ ha<sup>-1</sup> respectively. They explain these differences mainly by low-energy inputs in SRG biomass production and by the use of all above-ground biomass rather than just seed. These results have been considered as highly controversial and were strongly criticized (Russelle *et al.*, 2007). These comments themselves were discussed and the initial results were defended by their authors (Tilman *et al.*, 2007). The conclusions have to be extrapolated with care to European conditions notably because yields are different and C4 grasses are absent from European swards. Calculations from existing results and new experiments should be carried out in Europe. Tilman's experiment shows at least that energy balance calculations are not sufficient for comparing performances of different grasslands and crops, a lifecycle analysis taking into account carbon dynamic in soil (sequestration or release) and other GHG emissions is necessary in the context of GHG mitigation.

Still in North America, cropping perennial C4 grasses like switchgrass (*Panicum virgatum*) can produce 5 to 11 t DM ha<sup>-1</sup>, net energy yield of 60 GJ ha<sup>-1</sup> and 540% more renewable energy than non-renewable energy consumed (Schmer *et al.*, 2008). Lifecycle analysis showed that switchgrass crops are GHG-positive, neutral or negative depending on the use of inputs and subsequent yields. They have theoretically lower GHG emissions than annual crops notably because part of the biomass can be used for bio-refinery. Performances of *Miscanthus* crops should be checked in Europe.

In this new context of agricultural production, grasslands have advantages and shortcomings. They are summarized in a SWOT analysis of grasslands compared to other crops for agro-fuel production.

The strengths of grasslands are good DM yield and long persistency. Their lignin contents are low compared with some other second generation agro-fuels. They do not require pesticide use or only low amounts. The use of nitrogen fertilizer, high fossil energy demanding for its industrial synthesis, can be reduced by the presence of legumes in the sward. Their production costs are low (long persistency, nitrogen fixation). Nitrate and phosphate pollutions are reduced. They protect soil and water. Their effect on biodiversity could be positive or very positive especially with late cuts and lucerne. Some native and persistent legume and grass species (*Medicago sativa*, *Phalaris arundinacea*, *Arrhenatherum elatius*, *Dactylis glomerata*, *Festuca arundinacea*) and some exotic persistent grass species (*Miscanthus* spp.) are very well adapted to low-input biomass production in Europe. In contrast with trees (*Populus* spp. and *Salix* spp.), there is no interest to use GMOs in grassland. In contrast with first generation agro-fuels, marginal land could be used except for *Medicago*.

Weaknesses of grasslands are low sugar, starch and oil contents compared with first generation agro-fuels. In particular, methane production per kg biomass grass silage is low compared with maize silage. Abandoned or extensive grasslands are often located on sites where harvest of biomass is too expensive. An increasing demand for grassland biomass

could induce intensification (example: slurry spreading) of the whole grassland area including of species-rich swards. Grassland used for agro-fuel could have a positive effect on wildlife only with late cut systems, for instance one or two cuts per year while better energy yields seem to be achieved with several cuts. Apart from methane production, the technology for producing energy from ligno-cellulosic material like grasses and legumes are not yet mature. Its cost is thus higher than for producing energy from first generation agro-fuel.

The disappearance of the 'cereal and oilseed rape premium' of the former CAP system is an opportunity for grasslands. The protection of the grassland area is integrated in the present CAP cross-compliance system (GAEP). There is a social demand and political willingness to protect the environment, landscape and biodiversity. The increase of fossil energy and input prices can increase attractiveness of perennial grasses and legumes compared with first generation agro-fuels. An increase of cereal price can induce higher profitability of perennial species for agro-fuel production.

Threats can consist in a reduction of agricultural supports and a stagnation or a reduction of the importance of the rural development policy (second pillar) compared with the first pillar of the CAP. A significant proportion of the grassland area could be destroyed for production of first-generation agro-fuels. Agri-environmental premia could be reduced for finding financial resources of supporting agro-fuel production.

Cherney and Baker (2006) have developed a system of quotation of agro-fuels and applied it to grassland (Table 8). This table could be completed for first generation agro-fuels. It would certainly appear that they perform worse for GHG emissions, nutrient management benefits, soil conservation, specific societal benefits, pollution, pesticide usage and biodiversity.

Table 8. Scoresheet of environmental and production characteristics of grass agro-fuel (Cherney and Baker, 2006).

Criteria	Maximum score (including relative importance of each factor)	Grass biomass scores
Greenhouse gas emissions	15	14
Economic feasibility	10	9
Local energy security	9	9
Energy conversion efficiency	9	9
Nutrient management benefits	8	8
Soil conservation	8	8
Specific societal benefits	7	4
Pollution potential	6	4
Rural development	6	5
Grower acceptance	6	6
Land suitability	6	6
Maintaining open spaces	4	4
Pesticide usage	2	2
Wildlife nesting/winter cover	2	1
Encourage species diversity	2	1
Total	100	90

The same approach can compare the aptitudes of different grassland types for biomass production and GHG mitigation (Table 9). Apart from perennial C4 grass cropping, the best performing swards would be legumes and legume-based mixtures as well as extensive grasslands including species-rich swards.

Grassland biomass can be an energy source, but it may be additionally fractionated for the production of chemicals, biogenic materials and plant fibres. The 'Green Biorefinery Austria' is an example of the implementation of this idea (Wachter *et al.*, 2003). The raw material

from grassland (grass, legume or mixtures) is fully used to produce a multitude of 'product groups'. The 'Green Biorefinery' concept from Austria is different from other international projects in so far as it allows a continuous operation of the plant; it involves not only processing of direct-cut grass but mainly of grass silage that can be stored for year-round operation. The first stage of the process consists in a mechanical fractionation of the primary raw materials into a liquid (press juice) and a solid fraction (press cake). The liquid fraction contains valuable water-soluble substances (such as lactic acids and amino acids), the major part of the press cake consists of grass fibres. These fibres can be used for animal feeding, heating or fermentation in biogas plants. They should mainly be used as basic material for insulation materials (fibre boards, mats, fleeces, dry-blow injection insulation materials), building panels (chip boards, fibre boards, fireproof boards), materials for gardening and landscaping (mulch boards, plant bolsters, peat substitute), fibre reinforced composites (bio-composites, pre-formed parts for the automotive industry), packaging materials, additives for various building materials (bricks, plasters, mortars, wall fillers), gypsum fibre boards, paper and pulp, energy source (fuel pellets, biogas) and animal feed (pellets). The crude fibre yield from the press cake reaches 30% DM, which corresponds to about 95% of the total crude fibre content. Protein extraction seems not to be profitable yet but amino acids could be extracted from silage juice. They can be used for cosmetics, fermentation or food. Lactic acid produced by the ensiling process could be provided as a raw material for the chemical industry and could be used in the production of biodegradable plastics, environmentally friendly solvents and chemicals for the foodstuff industries. There is a wide market for this product. The technology has still to be developed but it is promising. It could use the grass produced in excess because of the decline of livestock and dairy farming, for instance the first spring cuts. It could have the effect of protecting cultural landscapes by stabilising the grassland area.

Table 9. Aptitudes of different grassland types for biomass production and GHG mitigation.

Grassland types	Aptitude for agro-fuel and GHG mitigation
Temporary	
Pure grass + N	---
Pure grass 0N	+ to +++
Grass + legume	+++
Persistent legume (lucerne)	+++
Permanent	
Species-poor, intensive +N	--
Species-poor, previously intensive, extensified 0N	+++
Species-rich, extensive 0N	++

---: very bad; --: bad; +: acceptable; ++: good; +++: very good.

## Discussion

The above data show that although the potential of forage production of grasslands is well documented, an important research effort has still to be devoted to the understanding of their potential for biomass and energy production.

Roeder *et al.* (2007) analysed the impact of the CAP reform of 2003 on the use of grassland in Europe on the basis of *ex-ante* analysis of this reform and of some more recent data. They concluded that decoupling would lead to a reduction in animal numbers (dairy and suckling cows, sheep). Accompanying measures like cross-compliance, the GAEC requirement and the implementation of the NATURA 2000 network would though stabilize the grassland area including in new Member States. They recorded also in recent years high price increases of cow milk and beef meat across several countries of Europe. This trend could reduce the decline in the number of animals. Many other factors make the predictions difficult. Climate

change mitigation policies could support the conversion of arable land to grassland for carbon sequestration; that could be implemented mainly in marginal cropping areas (South and North of Europe). The production of agro-fuels can have the reverse effect by increasing the competition of maize silage towards grass silage for methane production and of many crops (sugar beet, oilseed rape, cereals) towards grassland for ethanol and bio-diesel production. That could lead to a reduction of the grassland area and a further intensification of all grassland types including species-rich grasslands. The EU biomass action plan could though be reconsidered and the ambitions for agro-fuel production lowered following the pressure of some Members States like Germany, the OECD, the FAO, many stakeholders and even the World Bank and the IMF. Alternatively or in addition, a Code of GAEP could be imposed to first generation agro-fuel production during a transitory phase that would lead to the development of second generation agro-fuels. This code could impose the use of a minimum proportion of grassland biomass or the interdiction to produce crops for fuel on previous grassland areas. The increasing demand for cereals and other crops from developing countries could increase the pressure on the grassland area and encourage agricultural intensification in Europe. Price increases could also affect milk and meat which would temperate the decrease of grassland area but would be an additional encouragement for intensification. A reduction in the EU agricultural budget due to the enlargement and changes in the World trade policies could have contrasted effects in the grassland and livestock regions leading to extensification and abandonment in some regions and intensification in others. Table 10 summarizes the influence of several technical, economic, social and political factors on grassland systems components. It appears that the clearest trends are related with the increase of intensification of grassland and animal productions as well as food and feed prices. The changes in grassland area and animal numbers are more hazardous to predict.

Table 10. Influence of several technical, economic, social and political factors on grassland system components.

	Grassland yield	Grassland surface	Animal yields	Animal numbers	Feed prices	Meat and/or milk prices
Intensification	+	-	+	0		
Production specialisation	+	According to regions: + or - Average: -	+	-		
Milk quotas	+	0 or -	+	-		+
Decoupling	+	?		-		
		+ or - according to other factors				
Cross-compliance	0	0 (without: -)		0		
Modulation		+ (potentially)				
Nitrate Directive	0	0 or +	+	-		
Other environmental policies	0	0 or +		0 or -		
Reduction in beef and sheep meat consumption in Europe	0			-		
Increasing food demand from emergent markets		- or +?			++	+
Agro-fuels		--?			++	+

+: increase; -: decrease; 0: stable effect.

The future of grasslands and grassland-based systems cannot be separated from the multiple functions that they offer to society (see introduction). In this sense, grassland is not a crop like another and livestock systems are different from arable systems. Forage and livestock systems should be supported by the society for the services they provide including biodiversity conservation and landscape protection. They also offer recreational opportunities and they are

contributing to the quality of live by producing healthy and tasty products. That is an essential part of the European culture and heritage. A higher part of the CAP budget should be devoted to the second pillar of the CAP and should target grassland systems. Additionally, agricultural and environmental policies should be modulated according to the typology of livestock systems (Pflimlin *et al.*, 2005) in order to be better adapted to their specific needs. The EU is too large to apply the same policies from the North of Scandinavia to Malta and from Connemara to the Black Sea. It is easy to understand that dairy farmers from Scandinavia are facing different problems than shepherds from Sicily. Specific policy measures should be crossed with specific systems, even if a system type can include regions that are remote from each other in the EU: Spanish mountain farmers from the Pyrenees can have similar problems compared with Romanian farmers from the Carpathians. Grassland systems should be supported everywhere in the EU but the Mountain, Mediterranean and Grassland regions require the highest attention. The PDO policy of the EU should be reinforced, particularly in the countries in transition. These countries should be helped to implement this policy in regions where traditional systems were fought for about 50 years by centralized and very authoritative regimes. The agro-fuel policy of the EU should be reconsidered on the basis of the important amount of information that became recently available. Some political choices were obviously wrong and should be corrected. This concerns not only European citizens but also a big part of the World population that is undernourished or that could have increasing problems to find food because of the production of these agro-fuels. A new biomass policy must be defined for avoiding that the 'cure would be worse than the disease' (Doornbosch and Steenblik, 2007).

There is no doubt that the future increase of fossil fuel price and the consequent increase of the nitrogen fertilizer price will force European farming systems to a drastic reform in the next decades. In the future, systems will have to be less energy demanding. Fortunately, there are important possibilities to spare energy per ha or per ton of milk for instance (Haas *et al.*, 2001). Future researches have to focus systematically on energy costs and GHG emissions per production system and per product for developing such energy efficient systems. A new integration of grassland and arable land at the farm and/or the region levels will probably be necessary, for instance for reducing transportation costs. Nitrogen fixation by legumes will be one of the pillars of these future systems for saving the huge amounts of fossil energy that the artificial nitrogen fertilizers require. A special effort will have to be done by livestock systems because they are less energy efficient than arable systems per kg of food produced. Future systems will also have to release less GHG in the atmosphere, not only CO<sub>2</sub> but also CH<sub>4</sub> and N<sub>2</sub>O. Grasslands and grassland-based systems are thus facing a new revolution!

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# **Session 1**

## **Biodiversity and productivity in grasslands – strategies and limitations**



# Is the grass always greener on the other side of the fence? *Primula veris* L. as an example of plant survival at different management intensities

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## Abstract

The large number of threatened species, both animals and plants, in the European agricultural landscape has often been attributed to the abandonment of semi-natural grasslands. Species may, however, also be threatened in the remaining managed grasslands due to a changed and intensified management. In this study, effects of grazing management and population characteristics were examined in terms of their importance for the seed production and recruitment of the perennial herb *Primula veris* L. (cowslip). Four different life-cycle events related to reproduction were studied in around 30 populations, in 2003 and 2004, in an 11 km<sup>2</sup> landscape in Sweden, and related to a number of habitat and landscape properties. Population structure was examined in 11 populations in 2004. The loss of propagules in total and at each life-cycle transition was calculated. The results show that high proportions of propagules were lost due to grazing, unsuccessful pollination, and unsuccessful establishment of seedlings. No single explanatory variable could unambiguously explain unsuccessful pollination in both years. Grazed populations had, compared to ungrazed, on average lower seed set, higher seedling per seed ratio but equal density of seedlings, lower density of vegetative plants, and equal density of juvenile plants. Equal seedling density indicates that reduced seedling recruitment per seed in ungrazed grasslands was compensated by a higher production of seeds, and conversely, the loss of seeds by grazing in grazed areas was compensated by higher seedling recruitment per seed. The results from this study add to the increasing number of studies which indicate that low-intensity and site-specific grazing regimes have to be included in the management of semi-natural grasslands to maintain or enhance the diversity of plants and the diversity of organisms depending on the vegetation.

## Introduction

In Sweden one-third of the Red-Listed species are connected to the agricultural landscape, and a majority of these are found in semi-natural grasslands (Hansson and Fogelfors, 2000; Gärdenfors, 2005). The pattern is similar in all north-European countries; for example, in Finland, where more than 80% of the Red-Listed species in the agricultural landscape are linked to semi-natural grasslands (Luoto *et al.*, 2003). The loss of grassland area and the large numbers of threatened species raises the important question: how can we manage the remaining semi-natural grasslands in an optimal way?

The shift of management regimes throughout Europe's semi-natural grasslands is acknowledged as an important cause of the decline in grassland biodiversity (Wolking and Plank, 1981; Fuller, 1987; Stanners and Bourdeau, 1995). Intensified agriculture has caused degradation and abandonment of large grassland areas of Europe (e.g. Fuller, 1987; Luoto *et al.* 2003; Pykälä *et al.*, 2005), and intense and suboptimal management in others (Garcia, 1992; Beaufoy, 1998; Hansson and Fogelfors, 2000) leading to decreasing biodiversity in both cases.

To understand today's agro-ecological relationship it is essential to acknowledge the landscape history, as the main part of the semi-natural grasslands in Sweden, has a management history of several hundred years (Gustavsson *et al.*, 2007). In the 19th century the use of new techniques started to change the management of semi-natural grasslands, and

with the introduction of chemical fertilizers, essentially in the mid-20th century, the intensification of the landscape proceeded at a high rate. The traditional harvest of winter fodder in semi-natural meadows by mowing in summer (July to August), almost disappeared and was replaced by hay production on agricultural fields. The main area of semi-natural pasture, heterogeneous forest pasture, was transformed to ungrazed forest with intensified timber production and structural homogenization. Instead, animals were relocated to arable fields and to some extent to former semi-natural meadows (Gustavsson *et al.*, 2007).

The shift from traditional to modern land-use changed the prerequisites for a considerable number of species (Maurer *et al.*, 2006). It is possible to identify at least four important factors that species in semi-natural grasslands have had to face in the Swedish landscape during the last hundred years: a severe loss of habitat (Harrison and Bruna 1999; Berlin *et al.* 2000), changes of management type (Simán and Lennartsson, 1998), relocation of management types in the landscape (Gustavsson *et al.*, 2007), and a collapse of the structural and functional connectivity between grasslands (Eriksson *et al.*, 2002; Lindborg, 2006).

Many grassland species are persisting in the modern landscape but it is, even for non-threatened species, uncertain for how long. Few studies have described how populations are affected by the 'new management' of semi-natural grassland. Here we study a perennial grassland herb, *Primula veris* L. (cowslip), to examine how different management components affect different steps in the life cycle of a fairly typical grassland species.

Several studies show that litter accumulation may be negative for the persistence of grassland plant species, due to decreased seedling recruitment (Kelly, 1989; Huhta *et al.*, 2001; Ehrlén *et al.*, 2005). Grazing reduces litter depth, which increases the possibility for seedling recruitment but at the same time reduces seed production by removal of reproductive parts (Tamm, 1956; Grubb, 1977; Brys *et al.*, 2005). Additionally, grazing of flowers may cause other effects on plant populations, e.g. on pollination and the growth and survival of plants (Keasar *et al.*, 1996; Zimmerman, 1983; Vazquez and Simberloff, 2004).

Semi-natural grasslands are important nectar and pollen resources in many regions and may be crucial for the maintenance of pollinator populations (Westrich, 1996; Kruess and Tscharnke, 2002). The importance of pollinating insects for plant seed-set is well known (Holm, 1966), and are today one of the most used example of ecosystem services (Klein *et al.*, 2003; Ghazoul, 2005; Steffan-Dewenter *et al.*, 2005). Pollination is, however, not dependent on pollinator abundance alone, because pollinating insects may show selective foraging behaviour (Keasar *et al.*, 1996) in response to several factors, for example plant population size (Jennersten and Nilsson, 1993; Johnson *et al.*, 2004), plant population density (Sih and Baltus, 1987; Lennartsson, 2002; Kunin, 1993) plant species richness (Ghazoul, 2006), and presence of surrounding plant species (Thomson, 1978; Johnson *et al.*, 2003).

In this study, the loss of propagules in transitions between stages in the life cycle of *P. veris* was related to different characteristics of the populations, the sites and the landscape. The following questions were addressed: (1) to what extent are propagules lost at the different life stages under different conditions? (2) which environmental characteristics determine propagule loss? and (3) can population structure, i.e. the proportional relationship between different life stages, be explained by the effects on different life cycle events?

## Material and methods

### *Study area*

The approximately 11 km<sup>2</sup> study landscape consists of a mosaic of arable fields, pastures, and forests. It is situated on the Western part of the island Selaön in Lake Mälaren, in south-central Sweden (59°23'N, 17°09'E). The semi-natural grasslands in this part of Sweden have, in general, several hundred years of continuity as pastures or hay meadows.

The grassland patches in the landscape had, in most cases, unfertilized, uncultivated dry to mesic semi-natural vegetation. Abandoned grasslands are now in different stages of succession towards forests. The studied grasslands had the same management regime during the two study years (2003 and 2004) and during at least three years before the onset of the study.

### *Study species*

*Primula veris* was chosen as a study species because it has a synchronized flowering time, does not re-flower later in the season, it is self-incompatible and mainly reproducing by seeds (pollination is crucial), it flowers about two weeks after the usual onset of grazing in the area, it is decreasing in large parts of Europe (Brys *et al.*, 2004), and is an important host for several insect species.

### *Experimental design*

Thirty-one *P. veris* populations in 2003, and 28 populations in 2004, were included in the analysis of seed production. Eleven populations, three ungrazed and four grazed populations, were used in a population structure analysis and analyses of recruitment in 2004. For the reproduction study, plants were chosen randomly within the populations. Stalk height and number of flowers were noted for all plants. Factors that are presumed to influence population performance were: population size (area and numbers of flowering shoots), tree cover (measured as percentage of the population shaded by trees), litter depth (method explanation below), vegetation height (using a rising plate meter, Sanderson *et al.* (2001)), population isolation (mean distance to the two nearest populations), grassland isolation (area of grassland within 500 meters), flowering surface of *Salix caprea* and of other bee-pollinated plant species flowering at the same time as *P. veris* (estimated area of each tree or shrub within 200 meters), and management (grazing or not). Litter depth, from the mineral soil to the litter surface, was measured by using a ruler. Grassland area and grassland isolation were measured using aerial photographs and maps.

To examine the pollination, flowers in the same phenological stage were marked on the selected plants using acrylic paint, to avoid collecting flowers flowering at different days with different weather conditions, which may affect pollination. The fruits were collected before seed dispersal and the number of seeds, aborted seeds, and unfertilized ovules were counted in the lab.

Population structure was examined in five randomly distributed transects (20 cm wide and 2.7-12 meters long) per population. All *P. veris* individuals were counted and categorized as either: seedling juvenile plant, vegetative plant, or reproductive plant. Number of flowers per reproductive plant was noted.

Different life cycle events related to seed production and recruitment of *P. veris* were calculated to give the loss of propagules at each transition (Figure 1). A propagule was defined as a plant unit by which a new plant may be recruited; here: ovules, seeds, or seedlings. The studied environmental variables were population size, plant height, flowers per plant, litter depth, seed production, tree cover, grazing regime, area of adjacent grassland, mean distance to other populations, vegetation height, and surrounding floral richness before and during *P. veris* flowering. Two estimates were used to quantify propagule loss at each step: (1) total loss, i.e. the loss in relation to all possible propagules, i.e. ovules, and (2) transition loss, i.e. the loss at each transition from one stage to the next. The total loss may be interpreted as the probability of an ovule to be lost in a certain step, e.g the proportion of the potential number seeds (number of ovules) that are lost between dispersed seed to

seedlings. The transition loss is the probability of a propagule to be lost from one stage to the next, e.g. the proportion of seeds that are dispersed that are lost to the next stage.

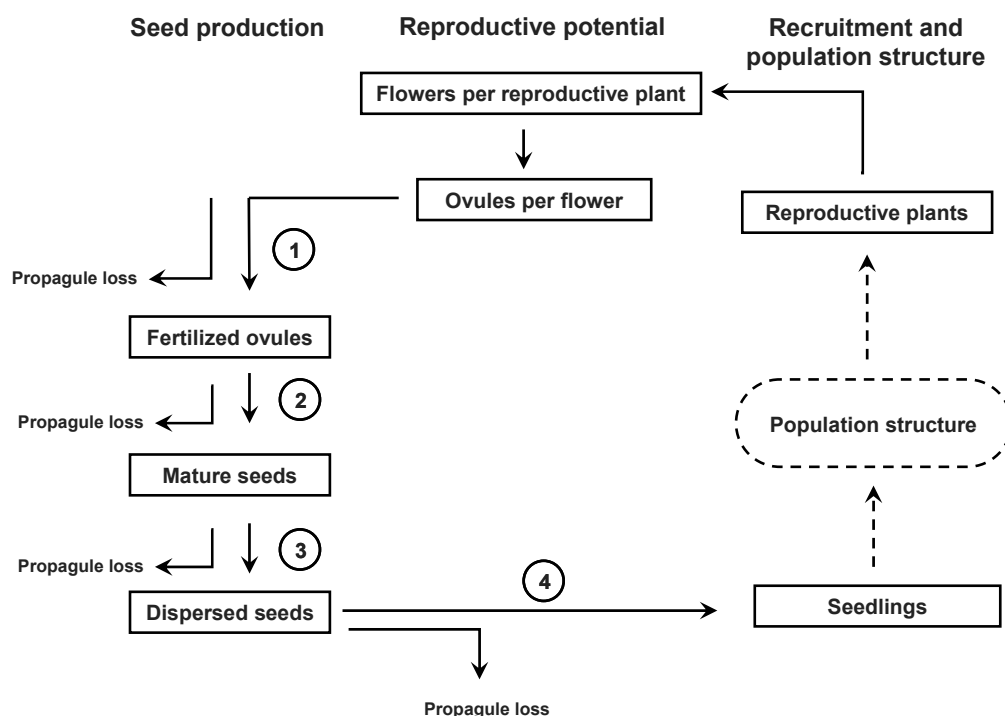


Figure 1. Different life stages related to the reproductive potential, seed production and recruitment of *Primula veris*. Transition numbers 1-4 represent propagule loss due to (1) insufficient pollination, (2) abortion of young seeds, (3) damage by grazers and herbivores, and (4) seed mortality or insufficient establishment of seedlings (see Table 2).

Table 2. Mean proportion (and one SE) of propagule loss in transitions between life stages according to Figure 1 in 2003 and 2004 (see text). The losses are calculated per propagule in the previous stage and per the total number of ovules. N = 31 in 2003 and 28 for transition 1-3 in 2004. N = 11 for transition 4.

Transition	Proportion of lost propagules							
	Per propagule in the previous stage				Per the total number of propagules (ovules)			
	2003		2004		2003		2004	
1	0.307	(0.042)	0.318	(0.028)	0.307	(0.042)	0.318	(0.028)
2	0.020	(0.006)	0.481	(0.014)	0.010	(0.002)	0.035	(0.010)
3	0.350	(0.051)	0.453	(0.050)	0.260	(0.052)	0.301	(0.042)
4*			0.987	(0.031)			0.418	(0.074)

\*Based on seed production 2003 and seedling number 2004.

### Statistical analyses

Multiple backward regressions were used to examine differences between populations and years in ovule number per flower and proportion of fertilized ovules. Each predictor variable, with a  $P < 0.4$  in univariate analyses, were retained and used in the multiple regression models (including all two-way interactions). Thus, no variables had to be excluded from the multiple regressions due to correlations among predictor variables.

The  $P$ -value for a variable to be kept in the backward regression was set to 0.05. Predictor variables were centred when necessary, to eliminate collinearity between interactions and lower order terms (tolerance for collinearity for all variables and analyses were higher than 0.20,



Quinn and Keough (2002)). The number of plants per population was the only variable which was transformed (log-transformation). All response variables, except the proportion of mature seeds per fertilized ovule, were normally distributed in histograms and quartile-quartile plots, and residuals from the fitted regressions were evenly distributed. All analyses were performed using Statistical Analysis System (SAS version 9.1).

## Results

Vegetation height was higher in ungrazed populations (mean  $\pm$  SE =  $13.1 \pm 1.1$ ) than in grazed populations (mean  $\pm$  SE =  $6.9 \pm 1.0$ ). Litter depth was positively correlated with vegetation height among populations (Linear regression  $R^2 = 0.69$ , DF = 1,  $F = 19.7$ ,  $P = 0.001$ ). Consequently, litter depth differed between ungrazed areas (mean  $\pm$  SE =  $4.0 \pm 0.38$ ) and grazed populations (mean  $\pm$  SE =  $1.28 \pm 0.22$ , one-way ANOVA,  $N = 11$ , DF = 1,  $F = 40.2$ ,  $P < 0.001$ ).

### *Reproductive potential*

The potential seed production of a plant in one season is determined by the number of flowers per plant and the number of ovules per flower (Burd, 1994). The number of flowers per plant differed between years, but not between populations (Table 1). In contrast, the number of ovules per flower differed between populations, but not between years. Populations were affected differently in the two years, indicated by a significant interaction between year and population (Table 1).

Table 1. Differences between populations and years in flowers per reproductive plant, ovules per flower, and proportion fertilized ovules (see text for explanations) in 29 populations of *Primula veris*.

Source	DF	Flowers per reproductive plant <sup>1</sup>		DF	Ovules per flower <sup>2</sup>		Proportion of fertilized ovules <sup>2</sup>	
		$\chi^2$	$P$		F	$P$	F	$P$
Year	1	31.7	< 0.001	1	0.1	0.705	2.5	0.113
Population	1	3.6	0.057	28	5.5	< 0.001	6.9	< 0.001
Population x Year	1	3.6	0.057	25	5.4	< 0.001	3.6	< 0.001

1 = Generalized linear model, Poisson distribution and log as link function, 2 = General linear model, ANOVA.

The number of ovules per flower was significantly associated with vegetation height in 2003 (multiple regression,  $R^2 = 0.38$ ,  $N = 27$ , DF = 1,  $F = 15.36$ ,  $P < 0.001$ ), but not in 2004. In 2004, the number of ovules per flower was instead mainly associated with the number of flowers per plant and plant stalk height ( $R^2 = 0.49$ ,  $N = 27$ , DF = 1,  $F = 8.1$ ,  $P = 0.003$ ; partial values: flowers per plant:  $t = 3.0$ ,  $P = 0.008$ , and stalk height:  $t = -3.2$ ,  $P = 0.005$ , DF = 1 for both variables). Only the number of flowers, plant stalk height, and vegetation height had a smaller  $P$ -value than 0.4 in the initial univariate test and were used in the full model. Thus, the reproductive potential (number of flowers per plant times number of ovules per flower) differed between years and was explained by different factors in the two years.

### *Pollination*

The proportion of fertilized ovules differed between populations and was significantly affected by the interaction population x year, but not by year alone (Table 1). None of the tested variables could explain differences between the populations in 2003 (best model  $P > 0.11$ ). In 2004 a significant negative relationship with the quantity of grassland within 500 meters was found ( $R^2 = 0.27$ ,  $N = 27$ , DF = 1,  $F = 5.9$ ,  $P = 0.027$ ).

### Abortion

The abortion of seeds after pollination was generally low but varied from 0 to 30% among populations and is therefore only potentially important for seed production in some populations. The abortion was positively correlated to the number of fertilized ovules in 2003 (Spearman rank correlation,  $N = 27$ , in 2003:  $r = 0.80$ ,  $P < 0.001$ , in 2004:  $r = -0.18$ ,  $P = 0.35$ ), but when a line was fitted to the significant correlation in 2003, the slope was close to zero, and thus the predictive value is questionable.

### Grazing and seed predation

A large proportion of the plants in grazed populations were grazed or trampled by cattle before the fruits matured, but plants, flowers, fruits and seeds were also lost due to effects of wild herbivores and seed predators (both mammals and insects, Figure 2). The different types of damage and their relative contribution to the total propagule loss was not possible to distinguish by the data, but the number of damaged plants was naturally much higher in pastures (mean  $\pm$  SE = in 2003;  $0.49 \pm 0.08$ , in 2004;  $0.27 \pm 0.07$ ) than in ungrazed grasslands (mean  $\pm$  SE = in 2003;  $0.09 \pm 0.03$ , in 2004;  $0.09 \pm 0.03$ ).

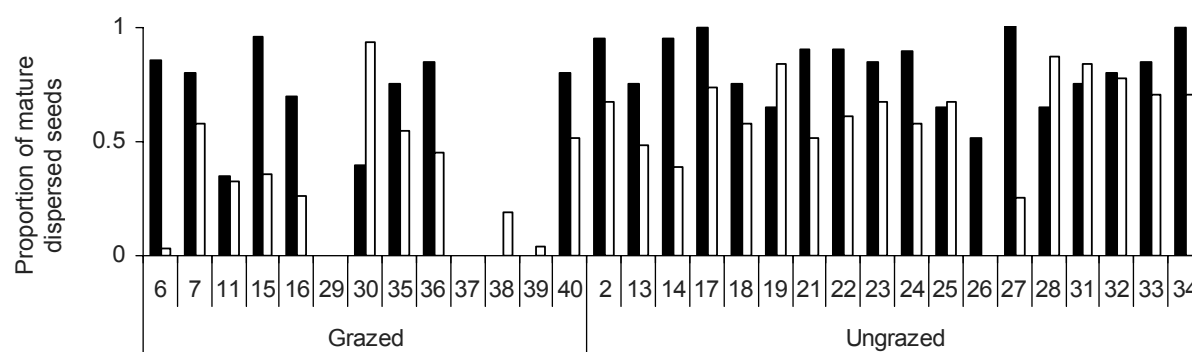


Figure 2. Population means for the proportion of mature dispersed seeds (seed set including grazing and predation) in 31 populations 2003 (closed bars) and 28 populations in 2004 (open bars). Values for population 29, 37, 38 and 39 in 2003 and population 29 in 2004 are zero, values for population 36 in 2003 and 26, 29, 36 and 37 in 2004 are missing. No error bars are shown because the values are based on mean values per population.

### Seedling recruitment

The number of seedlings per seed differed by a factor of 165 between populations and was significantly associated with litter depth (Figure 3).

### Relative importance of life cycle events

In 2004 most propagules were lost in the step between dispersed seed set to seedling, both as a single step and in relation to the total potential propagules (Table 2). Overall, only 1.3% of the produced ovules produced seedlings in the following year (Table 2). A high loss of propagules was also detected due to insufficient fertilization of ovules (Table 2). This study does not include detailed data on survival of seedlings to juvenile plants, but the proportion of juveniles per seedling was on average  $29 \pm 10$  % (mean  $\pm$  SE). Since juveniles may stay in the same stage more than one year, this is the maximum value of seedling survival (Brys *et al.*, 2005).

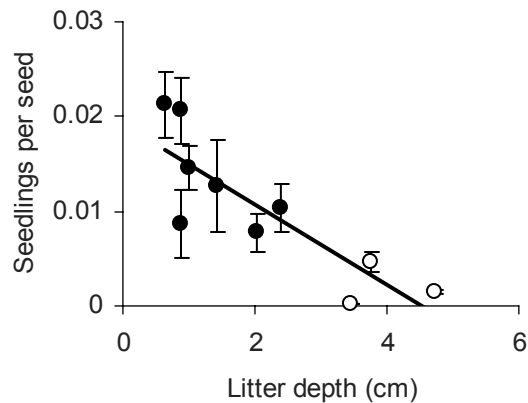


Figure 3. Seedlings per seed of *Primula veris* as a function of litter depth in 7 grazed (closed circles) and 3 ungrazed (open circles) populations in 2004. Linear regression,  $R^2 = 0.77$ ,  $N = 10$ ,  $DF = 1$ ,  $F = 27.0$ ,  $P < 0.001$ , one outlier was removed. Error bars show  $\pm$  one standard error.

#### Population structure

Grazed populations were generally larger (mean  $\pm$  SE =  $1836 \pm 1100$ ) than ungrazed populations (mean  $\pm$  SE =  $292 \pm 139$ ), but this may partly be due to differences in grassland size since small grasslands more often were abandoned than large ones.

Ungrazed populations in ungrazed areas had generally a larger proportion of the plants in adult stage than grazed populations (Figure 4). Despite differences in population structure between management regimes, neither seedling nor juvenile plant densities differed between grazed and ungrazed areas. In contrast, ungrazed populations had higher densities of both vegetative plants and reproductive plants (Figure 5).

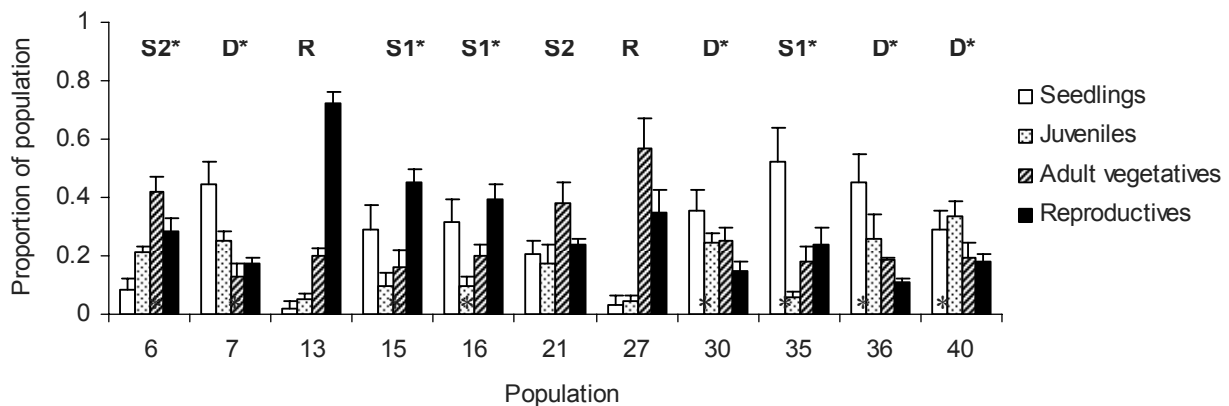


Figure 4. Proportion of life stages of 11 *Primula veris* populations. Asterisks indicate grazed populations and error bars show one standard error ( $N = 5$  transects).

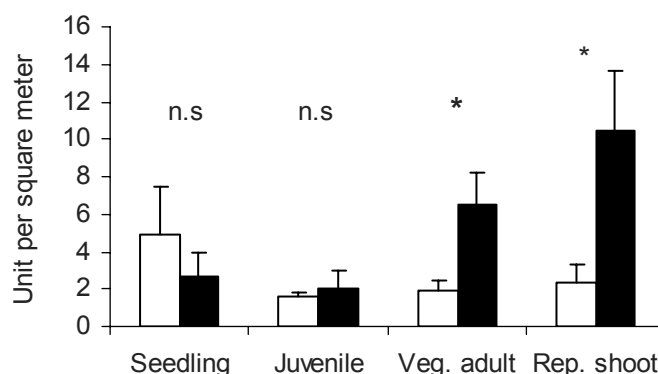


Figure 5. Density of seedlings, juvenile plants, vegetative adults and reproductive plants in 8 grazed (open bars) and 3 ungrazed (closed bars) in populations of *Primula veris*. Asterisks indicate significant differences (\* < 0.05, Kruskal-Wallis test).

## Discussion

This study on *P. veris* indicates that several different parallel management-related factors may influence the life cycle of a grassland plant in semi-natural grasslands, resulting in differences in reproductive output and population structure between populations. However, for the example-species, recruitment of new seedlings from seeds was the most sensitive stage, and was mainly determined by litter depth. Relatively large effects were also found on seed production, both due to pollination limitation and direct grazing of flowers and immature fruits. Landscape traits also influenced population performance, but these effects were not constant over time. Population structure was strongly influenced by management. Populations in grazed areas had high proportions of seedlings and juveniles, whereas ungrazed areas contained higher proportions of adult plants. Propagule loss due to grazing in grazed populations and low recruitment due to thick litter layer in ungrazed populations resulted in grazed and ungrazed populations having equal densities of seedlings and juvenile plants.

The potential seed production per flower is limited by the number of ovules, which was shown to vary among populations. Ovule number was associated with different variables in different years (positively affected by vegetation height in 2003 and positively by flower number but negatively by plant-stalk height in 2004). The differences between years indicate that other factors, not measured in this study, may also be associated with ovule number, for example light competition, nutrients or water resources (Burke *et al.*, 1998), or plant resource status and between-year differences in resource allocation to growth and seed production, respectively (Ehrlén and Eriksson, 1995).

Earlier studies have found pollen limitation in *P. veris* (Lehtilä and Syrjänen, 1995), while others have not (Garcia and Ehrlén, 2002). In this study, the proportion of fertilized ovules was low in several populations (range 0.2-1), and the transition from ovule to fertilized ovule accounted for the second highest loss of propagules. The proportion of fertilized ovules in 2004 was negatively correlated with grassland isolation whereas no correlations with any of the measured variables were significant in 2003. Patch isolation usually reduces pollination (e.g. Goverde *et al.*, 2002), although some studies have found that pollinators stay longer in isolated patches (e.g. Rasmussen and Brodsgaard, 1992). The difference between years may indicate that pollination is mainly a result of factors not studied here, such as between-year variation in pollinator abundance, or site-specific climate differences.

The abortion of young seeds or fertilized ovules is not commonly considered in pollination studies, but was in this study found to be relatively high (up to 30%) in some of the populations. The abortion was correlated with the number of fertilized ovules in 2003, which

may indicate resource deficit, but the correlation-slope was near zero, indicating that the variation in abortion rate is of little ecological significance.

Grazing cattle and to some extent herbivory by insects were responsible for the loss of about a third of the potential propagule production, which naturally was the highest in grazed populations. In addition to the direct removal of reproductive parts, intense grazing may affect plants by damage of leaves or roots by trampling, which may affect future plant performance (Ehrlén *et al.*, 2005).

Of the seeds that survived grazing, a majority (on average 98.7%) died between dispersal and the establishment of seedlings. This was largely explained by litter depth, and the relation between litter depth and seedlings per seed indicated a threshold for seedling recruitment at about four centimetres. Litter depth was associated with vegetation height, which in turn was determined by grazing intensity. Thus, grazing promoted higher seedling establishment, but the seed production was reduced by grazing.

The population structure differed between populations and was associated with management. High proportions of young plants may be due to either high recruitment of new plants or high mortality of old plants. Despite the higher seedling per seed ratio in grazed areas, neither seedling nor juvenile density differed between managements. The density of reproductive plants was higher in ungrazed areas which compensated for the lower recruitment. High seedling densities but low total densities in grazed populations indicates rapid population turnover. This may be expected since disturbance of trampling and grazing of plant has been shown to have negative effects on adult plant survival (Garcia and Ehrlén, 2002).

### *Implications for conservation*

Grassland species, and among them *P. veris*, are decreasing in many parts of Europe (Kery *et al.*, 2000, Brys *et al.*, 2004). The decrease is often attributed to habitat destruction through ceased management. But, this study suggests that intense grazing may have similar effects on populations as ceased management, as long as the grassland persists in an open stage (Luoto *et al.*, 2003; Pykälä *et al.*, 2005). Grazed populations may suffer from low seed set and high mortality of adult plants, whereas ungrazed populations suffer from low recruitment rate. The results thus suggest that the most favourable management regimes for *P. veris* are those that remove litter but favour seed set. The phenology of *P. veris* indicates that this species, similar to many other grassland plants (e.g. Lennartsson and Oostermeijer, 2001; Brys *et al.*, 2005; Ehrlén *et al.*, 2005) is favoured by late-season management, e.g. late summer mowing. But, management by mowing is time consuming and may not be suitable for most areas with intensified and highly mechanized agriculture. Therefore, it is crucial to find new solutions to meet biodiversity's requirements for managements that are adjusted to ecological criteria at the same time as they are economically profitable. Also low-intensity grazing or variation in management intensity, i.e. high recruitment some years and high survival in others, should promote population growth. This may be achieved by every second or third year without grazing (Wissman, 2006). Such low intense, variable or delayed management regimes would also favour other grassland organisms dependent on nectar, pollen, seeds, or undamaged plants.

This study shows similar outcomes in abandoned grasslands and intensely grazed pastures, which indicate that the modern management may not be enough to conserve species with similar ecology. Several declining species show similar phenology as *P. veris*, but some of them are short lived and react more rapidly to changes in management. Therefore, the threat to long-lived species and ecological systems connected to them may be underestimated. Long-lived species may persist in harsh environment a long time after that the favourable conditions derived through a precise management have ceased, so-called extinction debt. The high numbers of Red-Listed species in the agricultural landscape indicate that the change in land

use during the last 50 to 100 years may have caused an extinction debt that is now made visible by the ongoing decline of grassland species, also in managed grassland habitats. Studies like this may help in detecting threats to plant populations by identifying suboptimal management regimes that constitute a long-term threat to species that are still common.

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# The benefits of sward diversity for cultivated grasslands

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## Abstract

Increased emphasis on the multifunctionality of European agriculture has called for a re-investigation of the use of mixtures in productive grasslands. Recent ecological research, carried out in relatively species-rich and nutrient poor systems, has indicated that ecological processes may be more effective when species diversity increases. If this would also be valid under nutrient-rich intensively managed conditions, increased crop diversity in species-poor agronomic systems could improve the provision of ecosystem services. We have recently developed an experimental and modelling framework which allows us to separately estimate the contributions of interactions in species mixtures by manipulating relative abundance. We applied our methodology to data from a common experiment at 28 European sites, using mixtures consisting of four agronomic species belonging to four functional groups. Our results indicate strong benefits of sward diversity in intensive cultivated grassland systems. Generally, mixtures provided more forage yield than could be expected on the basis of the performance of all species in monoculture. Averaged across sites we found that the performance of mixtures generally even exceeded that of the best performing monoculture. Mixtures strongly reduced the incidence of unsown species in the sward. The diversity effect was consistent over a huge range of environmental conditions, thus adding generality to our findings. It appears to persist over three harvest years and even under very intensive management. In contrast to other biodiversity experiments, our methodology has allowed us to determine the significance of species' evenness for diversity effects.

Keywords: agricultural grasslands, grass-legume mixtures, yield stability, plant-species diversity, overyielding

## Introduction

In recent decades, productive grasslands in many European countries have developed towards pure stands, here after referred to as monocultures, of single grass species with high yield potential and feeding value, maintained by high levels of nitrogen and frequent harvesting and/or high stocking rates (Wilkins *et al.*, 2002). Some countries, though, have continued to use mainly grass-clover mixtures with four to eight species (e.g. Frey, 1955; Suter *et al.*, 2004; Frankow-Lindberg, 2005) even on intensively managed grasslands, and Caradus *et al.* (1996) call white clover 'the key to the international competitive advantage of New Zealand's

pastoral industries'. There remains, however, a need to systematically test monocultures and mixtures under a range of environmental conditions and management intensities, to test the effect of the relative abundance of the species in a mixture, and, consequently, test which mixture composition produces largest diversity effects.

As in any ecosystem, agricultural systems are dependent on a variety of biological processes that arise from the activities of biological organisms. Grasslands are composed of species whose activities provide services that benefit society, e.g. provision of food, forage, fibre and fuel. At another scale, agricultural ecosystems are also dependent on a host of other biological organisms in the soil, in adjoining habitats that in turn provide services such as decomposition, maintenance of soil fertility, provision of clean water, pollination etc. Recent ecological research investigates the extent to which the provision of such ecosystem services is dependent on species diversity. In general, such research has been conducted by ecologists in relatively species-rich and nutrient-poor systems, and it seems that ecological processes may be more effective when species diversity increases (Tilman *et al.*, 1996). If this was also valid under nutrient-rich, intensively managed conditions, increased crop diversity in species-poor agronomic systems could improve the provision of ecosystem services (Hooper *et al.*, 2005). However, such predictions are largely untested due to the rarity of multi-species agronomic experiments that use more than two species, many of which have been legume-grass or legume-crop combinations (Vandermeer, 1989; Federer, 1999; Gibson *et al.*, 1999). Sanderson *et al.* (2004) reviewed results from forage productivity trials with diverse mixtures, and in a number of trials there were indications that productivity of pastures increased with increasing numbers of productive species. This was, though, not true for all trials reviewed making it difficult to draw general conclusions.

It is difficult to determine, both in principle and practice, how many species are needed to give maximum diversity effects (Hector and Loreau, 2005). It has been suggested that a highly species-rich system may not necessarily meet the primary objective of producing high yields in a highly productive and stable environment, whereas it may be advantageous in highly heterogeneous environments serving a multifunctional role (Sanderson *et al.*, 2004). In the former case the diversity effect would not rely on highly species-rich communities (> 30 species) but could be obtained with a mixture of few species well adapted to the appropriate environmental conditions.

It has been suggested that maintaining an even distribution of competitive forage species in the mixture, rather than the forage species number, would be important for diversity effects (e.g. reducing weed invasion, Tracy *et al.*, 2004). However, studies on diversity and ecosystem functioning have generally used experimental designs that ignore or confound important components of diversity (relative abundance, richness and identity) as discussed by Connolly *et al.* (2001) and Schmid (2002). Most of these studies centre on the number (richness) or composition of species or functional groups in the community ignoring the active determinant of relative abundance (evenness) of species. Where the effect of diversity depends on interspecific interactions (e.g. Harper, 1977), it has been pointed out that the contribution of a particular interaction depends on the strength of the interaction and the relative abundance of the species involved (Sheehan *et al.*, 2006). The net effect of interactions may lead to overyielding, where the performance in mixture exceeds that expected from monoculture performance, or even to transgressive overyielding (Trenbath, 1974), where mixture performance exceeds that of the highest yielding monoculture.

We have recently developed an experimental and modelling framework which allows us to separately estimate contributions of interactions in species mixtures by manipulating relative abundance (Kirwan *et al.*, 2007). By applying this framework in an experiment across a number of sites in Europe, using mixtures consisting of four agronomic species belonging to four functional groups, we are able to address such basic questions in agronomic diversity-function research as: (i) can positive interactions between important agronomic species result in greater yields from mixtures than expected from the performance of the individual species sown as monocultures; (ii) can such benefits persist over several years; (iii) can species diversity prevent invasion of unsown species into the swards; (iv) are diversity effects consistent across large spatial scales; (v) are diversity effects consistent under highly fertilized conditions?

### **Description of the COST 852 Agrobiodiversity Grassland Experiment**

Under the auspices of COST 852 a common experiment was carried out at over 40 sites in Europe and at one site in Canada (Helgadóttir *et al.*, 2005) during 2002-2007 with the primary aim of quantifying diversity-function relationships in agro-ecosystems. The experimental design allows separate analysis of the effects of the components of diversity – species relative abundance (evenness), richness (number) and identity – on community function and structure in a range of four-species (representing different functional groups) mixtures with widely varying relative species proportions and monocultures sown at two densities (Kirwan *et al.*, 2007).

#### *Experimental design, sites and measurements*

At each experimental site adapted cultivars of two legume and two grass species were chosen such that one of the grass and one of the legume species was fast establishing and the other was slow establishing. Five species-groups were selected depending on the geographical region of the experimental site: north European (NE), mid-European (ME), dry Mediterranean (DM), moist Mediterranean (MM) and a fifth group (Other) consisted of sites, each with its own group of species but all with the same four functional groups (Kirwan *et al.*, 2007). The experimental layout followed a simplex design (Cornell, 2002) with four monocultures and 11 mixtures of the four species sown at two levels of overall sowing density (low being 60% of high) (Figure 1). The 11 mixtures consisted of four mixtures dominated in turn by each species (sown proportions of 70% of dominant and 10% of each other species), six mixtures dominated in turn by pairs of species (40% of each of two species and 10% of the other two) and the centroid community (25% of each species). Species proportions at sowing were based on proportions of seed mass considered appropriate for monocultures for each species at a site. The experimental design allowed for adding extra plots testing selected factors of interest at a site, such as levels of N fertilization, genetic diversity of the legume component and cutting frequency.

A common protocol was established for plot management. The plots were harvested three to five times per annum, as appropriate for local conditions, for three years. Annual application of nitrogen fertilizer ranged from 0 to 200 kg ha<sup>-1</sup> in the basic plots and was extended up to 450 kg ha<sup>-1</sup> in additional plots testing N fertilization effects. For estimation of total annual above-ground biomass and botanical composition, including unsown species, a subplot was cut to a height of 5 cm at each harvest. At a number of sites additional parameters have been measured for the different communities such as soil microbial and arthropod diversity, nitrogen economy (N-content of herbage, N<sub>2</sub>-fixation, gaseous N losses, N-leaching) and quality of harvested biomass.

Here we will present results of the first harvest year from 27 sites across Europe (Figure 2). In addition we present results from preliminary analysis of three complete harvest years from 12 out of the 15 sites using the ME species mixture (Table 1).

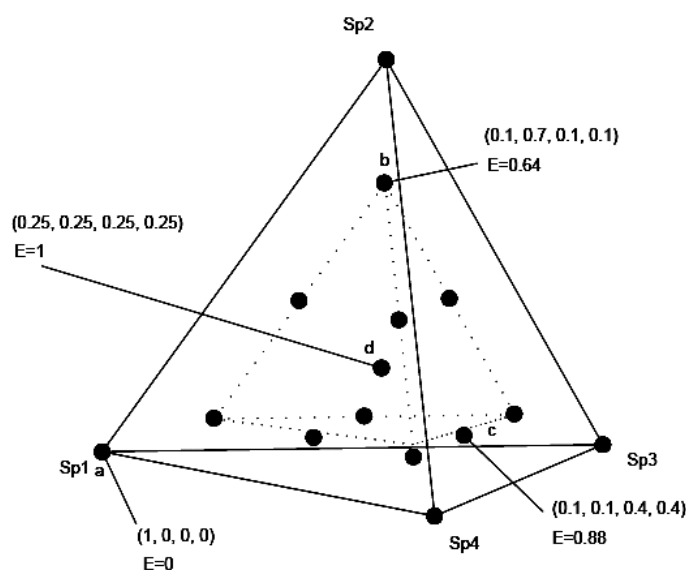


Figure 1. Graphical representation of the four species simplex design. Each point in the tetrahedron represents a community with its position determined by its sown relative abundance pattern (Sp1, Sp2, Sp3, Sp4). The 15 experimental communities in our design consisted of four monocultures (e.g. a), four mixtures dominated in turn by each species (e.g. b), six mixtures dominated in turn by pairs of species (e.g. c) and the centroid community where all species are equally represented (d). (Taken from Kirwan *et al.*, 2007).



Figure 2. Location of the experimental sites displayed according to the geographical species-groups: Mid-European (ME, ×), Northern-European (NE, ■), Moist-Mediterranean (MM, △), Dry-Mediterranean (DM, □) and the sites that used their own site-specific species (Other, ●). (Taken from Kirwan *et al.*, 2007).



### Data analyses

Species contribute to ecosystem function in different ways and we account for these species-specific identity effects (ID) in our analysis based on Kirwan *et al.* (2007). A diversity effect (D) is the excess of mixture performance over that expected from the component species identity effects. We model a functional response (EF) as a function of identity and diversity effects ( $EF = ID + D$ ).

The first component (ID) is modelled as a simple function of initial species proportions in the seed mixture.

$$ID = \sum \beta_i P_i$$

where  $P_i$  is the initial sown proportion of species  $i$  and  $\beta_i$  is its identity effect. Were there no diversity effects, the ecosystem function of a mixture would be a weighted average of the component species identity effects. Diversity effects are produced by interactions among the species in a mixture.

$$D = \sum \delta_{ij} P_i P_j$$

where  $\delta_{ij}$  is the pairwise interaction among species  $i$  and  $j$ . The  $\delta_{ij}$  may be positive indicating complementarity or facilitation, or negative indicating antagonistic effects. There may be patterns among the  $\delta_{ij}$  relating to functional groups, etc., or interactions may be more complex involving more than two species. The D component of the model generalizes naturally into a hierarchy of models that allows testing among alternative descriptions of the diversity effect. All pairwise interactions may be of similar sign and magnitude. When this is the case, the diversity effect may reduce to a simple function of initial community evenness (E).  $D = \delta E$ , where  $\delta$  is the average interaction strength. In Kirwan *et al.* (2007) we found that an evenness model was the best description of the diversity effect when we analysed above ground biomass in the first year across 28 sites. We are extending this model to test whether diversity effects persist into the third year and are including random effects to allow for site to site variation in the model coefficients and also repeated measures variation across time. Our modelling approach, based on initial sown proportions and densities of species, will predict the long term outcome of particular planting schemes; explanation of annual outcomes in terms of the dynamics of systems over years awaits further analysis.

### Sward diversity and biomass production

Our data showed consistent positive effects of diversity on total yield of plots originally sown with an equal proportion of all species (centroid) (Table 1) and it is remarkable that these positive effects of diversity were consistent over a range of productivity from 1.0 to 13.6 t ha<sup>-1</sup> and a spatial range from Spain to Iceland.

Preliminary analysis of the 12 sites using the ME species-group shows that the diversity effects persisted in the second harvest year at virtually all sites (Table 1). The same holds true for the third harvest year (data not shown). In a combined analysis across the 12 sites the mean total yield of the mixtures was on average 33-39% higher than average monoculture yield and when only the yield of the sown species was considered (omitting unsown species) the diversity effect was much greater (45-95%) and it increased with time. When looking at results over three harvest years the yield of the individual species sown in monocultures reduced with time, particularly for the fast establishing species, and unsown species contributed progressively more to the total yield on these plots (Figure 3). The mixtures at the centroid, on the other hand, maintained their advantage in total yield over the monocultures throughout and ingress of unsown species was minimal. By the third harvest year around 65% of the yield on plots sown originally with *Trifolium pratense* in monoculture consisted of

unsown species and only plots with *Dactylis glomerata* contained less than 10% unsown species. At the same time unsown species made up around 3-6% of the total yield of the mixtures.

Table 1. Estimates of the diversity effect in mixtures at the centroid for total yield, including unsown species (tonnes DM ha<sup>-1</sup>), at 27 sites in harvest year 1 and 17 sites in harvest year 2 (coefficients in **bold** indicate significance at  $\leq 5\%$ ). Also shown for each site in year 1 are species groups and mean monoculture yield. The sites included in the preliminary analysis over three harvest years are shown in *italics*. (Taken from Kirwan *et al.*, 2007).

Country	Species-group	Mean monoculture yield (t DM ha <sup>-1</sup> )	Estimated diversity effect at the centroid D $\pm$ SE (t DM ha <sup>-1</sup> )	
			Year 1	Year 2
<i>Germany</i>	ME	13.6	<b>4.84</b> $\pm$ 0.835	
<i>Ireland</i>	ME	13.6	<b>3.75</b> $\pm$ 0.515	<b>1.51</b> $\pm$ 0.641
<i>Lithuania (a)</i>	ME	5.5	0.34 $\pm$ 0.545	<b>1.84</b> $\pm$ 0.543
<i>Lithuania (b)</i>	ME	8.7	<b>2.44</b> $\pm$ 0.681	
<i>Lithuania (c)</i>	ME	9.1	<b>2.17</b> $\pm$ 0.464	
<i>Netherlands</i>	ME	8.4	<b>3.70</b> $\pm$ 1.069	<b>5.44</b> $\pm$ 1.375
<i>Norway (a)</i>	ME	7.8	<b>6.85</b> $\pm$ 0.602	<b>4.08</b> $\pm$ 0.402
<i>Norway (b)</i>	ME	9.9	<b>2.13</b> $\pm$ 0.36	<b>3.44</b> $\pm$ 0.579
<i>Poland</i>	ME	6.7	<b>1.77</b> $\pm$ 0.489	<b>2.66</b> $\pm$ 0.610
<i>Spain (a)</i>	ME	6.8	2.01 $\pm$ 1.217	
<i>Sweden (a)</i>	ME	7.9	<b>2.84</b> $\pm$ 0.582	<b>5.75</b> $\pm$ 0.548
<i>Sweden (b)</i>	ME	7.2	<b>3.72</b> $\pm$ 0.496	
<i>Switzerland</i>	ME	10.4	<b>5.64</b> $\pm$ 0.689	<b>7.46</b> $\pm$ 0.573
<i>Wales (a)</i>	ME	6.7	<b>4.37</b> $\pm$ 0.583	<b>2.21</b> $\pm$ 0.674
<i>Wales (b)</i>	ME	6.8	<b>4.68</b> $\pm$ 0.655	<b>4.05</b> $\pm$ 0.729
<i>Iceland (a)</i>	NE	4.3	<b>1.13</b> $\pm$ 0.533	0.49 $\pm$ 0.293
<i>Iceland (b)</i>	NE	1.3	<b>1.16</b> $\pm$ 0.262	<b>1.25</b> $\pm$ 0.239
<i>Norway (c)</i>	NE	7.1	<b>4.18</b> $\pm$ 0.481	
<i>Norway (d)</i>	NE	8.1	<b>2.40</b> $\pm$ 0.465	<b>2.04</b> $\pm$ 0.334
<i>Sweden (c)</i>	NE	7.2	<b>2.38</b> $\pm$ 0.32	
<i>France</i>	MM	8.1	1.71 $\pm$ 1.002	
<i>Greece</i>	MM	2.5	<b>0.92</b> $\pm$ 0.222	
<i>Italy (a)</i>	MM	8.0	<b>1.50</b> $\pm$ 0.712	
<i>Italy (b)</i>	DM	1.9	<b>1.62</b> $\pm$ 0.217	<b>0.91</b> $\pm$ 0.428
<i>Spain (b)</i>	DM	1.0	<b>1.70</b> $\pm$ 0.263	<b>3.57</b> $\pm$ 0.684
<i>Belgium</i>	Other	11.6	<b>5.18</b> $\pm$ 0.577	<b>9.14</b> $\pm$ 0.789
<i>Denmark</i>	Other	9.7	<b>4.57</b> $\pm$ 0.488	
<i>Finland</i>	Other	7.0	<b>2.65</b> $\pm$ 0.355	

Preliminary analysis of the 12 sites using the ME species-group shows that the diversity effects persisted in the second harvest year at virtually all sites (Table 1). The same holds true for the third harvest year (data not shown). In a combined analysis across the 12 sites the mean total yield of the mixtures was on average 33-39% higher than average monoculture yield and when only the yield of the sown species was considered (omitting unsown species) the diversity effect was much greater (45-95%) and it increased with time. When looking at results over three harvest years the yield of the individual species sown in monocultures reduced with time, particularly for the fast establishing species, and unsown species contributed progressively more to the total yield on these plots (Figure 3). The mixtures at the centroid, on the other hand, maintained their advantage in total yield over the monocultures throughout and ingress of unsown species was minimal. By the third harvest year around 65%

of the yield on plots sown originally with *Trifolium pratense* in monoculture consisted of unsown species and only plots with *Dactylis glomerata* contained less than 10% unsown species. At the same time unsown species made up around 3-6% of the total yield of the mixtures.

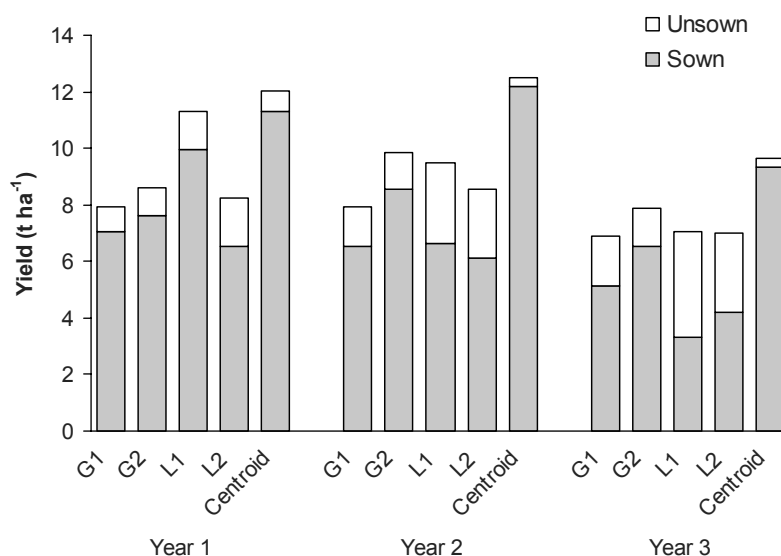


Figure 3. Predicted sown and unsown yield ( $\text{t DM ha}^{-1}$ ) for each monoculture and the centroid mixture for each year from the combined analysis across 12 ME sites. The unsown yields in this diagram are computed as the difference between estimated means of Total yield and Sown species yields. G1: *Lolium perenne*, G2: *Dactylis glomerata*, L1: *Trifolium pratense*, L2: *Trifolium repens*.

In harvest year 1 the diversity effect for the ME species-group showed a clear saturation with increasing evenness. This suggests that a major diversity benefit for yield in cultivated grasslands could be achieved with as few as four agronomic plant species in this system (Figure 4). Preliminary analyses indicate that similar results were obtained for harvest years 2 and 3. In natural systems there may be additional effects of diversity at higher levels of species richness (Hooper *et al.*, 2005).

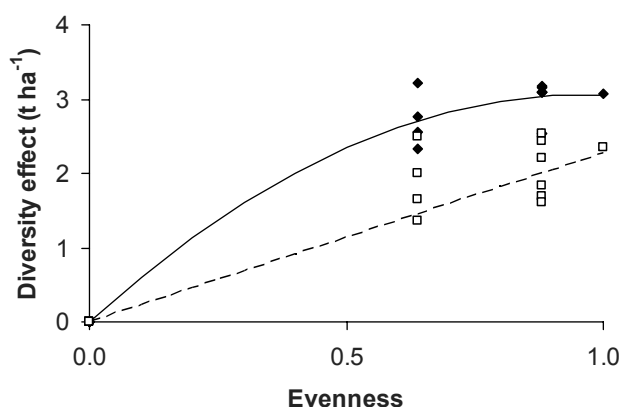


Figure 4. Average diversity effects ( $\blacklozenge$  for ME and  $\square$  for NE) and predicted diversity effects (— for ME and --- for NE) for harvest year 1. Average diversity effects for the 11 mixed communities were computed as the mean yield for the community minus the weighted average of the monoculture yields, weighted by the sown species proportions in the community. Models are based on sown proportions of species. (Taken from Kirwan *et al.*, 2007).

There were clear indications of transgressive overyielding for most of the mixtures in the first harvest year (Figure 5) and preliminary analysis of the 12 sites with the ME species-group indicated that all mixtures yielded significantly more than the highest yielding monoculture in subsequent years. This was estimated as 6%, 20% and 16% for harvest years 1-3, respectively, based on comparing the average total mixture yield with the highest yielding monoculture. If only sown species yields were considered the effects would be even greater.

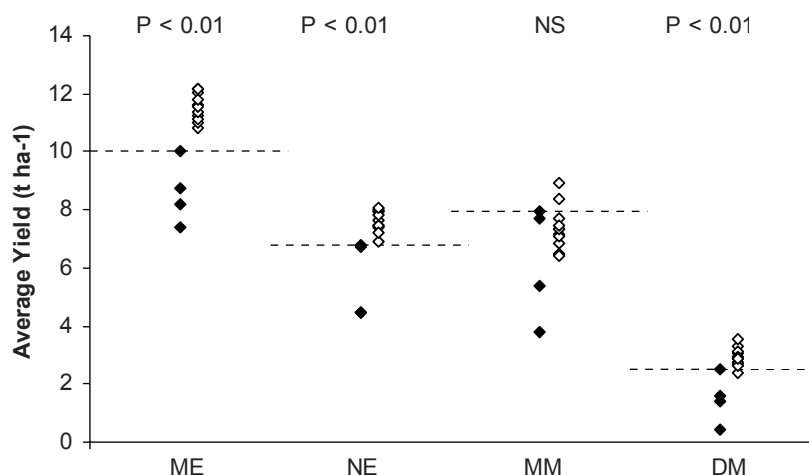


Figure 5. Average total yield (including unsown species) across sites for the four different species-groups of the four monocultures (♦) and 11 mixture communities (◇) for harvest year 1. (Taken from Kirwan *et al.*, 2007).

### Can mixtures contribute to sustainable agriculture?

It is generally considered that increased plant species richness, and particularly functional group richness, leads to greater biomass production in nutrient poor grassland systems (Hooper *et al.*, 2005). At the Swiss site of the common experiment (Zürich-Reckenholz) it was investigated whether this diversity-productivity relationship holds for high levels of nitrogen averaged over three harvest years (Nyfeler *et al.*, 2008). It was found that overyielding of the centroid mixture was 62%, 55% and 47% at 50, 150 and 450 kg N ha<sup>-1</sup> yr<sup>-1</sup> respectively. Even though overyielding decreased with increasing N fertilization, transgressive overyielding was still observed at the very high N fertilization of 450 kg N ha<sup>-1</sup> yr<sup>-1</sup> (Figure 6). It is remarkable that mixtures receiving low N input (50 kg N ha<sup>-1</sup> yr<sup>-1</sup>) could produce similar forage yields as the highest yielding grass monoculture receiving the very high N level of 450 kg N ha<sup>-1</sup> yr<sup>-1</sup> (Figure 6). These results demonstrate that high productivity can be maintained and large quantities of N fertilizer can be saved by the use of well balanced mixtures in intensive grassland systems.

The common experiment has given us the opportunity to take a broader whole-system view on ecosystem processes, such as studying the effects of species diversity on trophic interactions (e.g. Roscher *et al.*, 2004). Thus, the effects of plant diversity on arthropod communities were investigated at two sites (Johnstown Castle, Ireland and Gòsol, Catalan Pyrenees). Preliminary results indicate that there were significant positive effects at both sites of plant diversity and evenness on arthropod species richness and abundance, both for the total arthropod responses and for the detritivore and parasitoid groups (Llurba *et al.*, 2007). In Johnstown Castle, it was found that evenness explained a part of the variability not explained by yield for the total and parasitoid species richness and abundance, as well as for predator

species richness and detritivore abundance. These results therefore indicate evidence in support of cascading effects of diversity up the food chain.

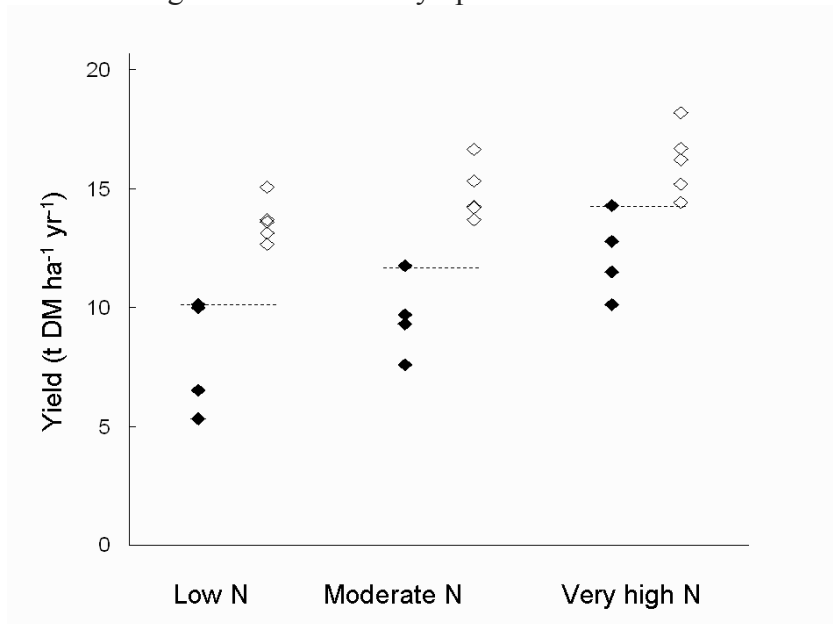


Figure 6. Average total yield (including unsown species) of monocultures (◆) and mixture communities (◇) grown at Zürich-Reckenholz, Switzerland at three levels of N fertilization (means over harvest years 1-3). Low N: 50 kg N ha<sup>-1</sup> yr<sup>-1</sup>; Moderate N: 150 kg N ha<sup>-1</sup> yr<sup>-1</sup>; Very high N: 450 kg N ha<sup>-1</sup> yr<sup>-1</sup>. (From Nyfeler *et al.*, 2008).

## General conclusions

The results from the COST 852 Agrobiodiversity Grassland Experiment presented in this paper indicate strong benefits of sward diversity in intensive cultivated grassland systems already at a low species number. The same holds true for extensive grassland located under sub-optimal environmental conditions (e.g. in rainfed Mediterranean regions). Mixtures provided more forage yield than could be expected on the basis of the performance of all species in monoculture. Averaged across sites we found that the performance of mixtures generally even exceeded that of the best performing monoculture. Mixtures strongly reduced the incidence of unsown species in the sward. It is, of course, well known that grass-legume mixtures can give yield advantage compared to monocultures (Boller and Nösberger, 1987; Spehn *et al.*, 2005; Zanetti *et al.*, 1997). However, the diversity effect was much greater than expected from these studies and, as our analyses indicate, it was not only a legume effect as the positive interaction between two grass species and two legume species was as strong as that between a grass and a legume (Kirwan *et al.*, 2007). The diversity effect was consistent over a large range of environmental conditions, thus adding generality to our findings, and it appears to persist over three harvest years and even in very intensively managed systems.

These results and increased productivity with increasing species richness observed in nutrient-poor systems appear to contradict conventional understanding about agricultural swards, subjected to intensive management and high fertilizer levels, in which monocultures or bi-species mixtures are often recommended. Such intensively managed agroecosystems on fertile soils are faced with much less environmental heterogeneity and nutrient and water limitation than nutrient-poor ecosystems and light becomes the limiting resource for plant growth. Competition for light is a positive feedback process, favouring the dominance of fast growing species that convert light energy into aboveground biomass most efficiently (Schulte *et al.*, 2003) at the expense of slow growing species adapted to nutrient-poor conditions. This model

is in line with the hump-back model (Grime, 1973) predicting competitive exclusion and, thus, strongly declining species richness with increasing fertility and productivity. This has, indeed, been confirmed in many studies (e.g. Wilson and Tilman, 2002; Crawley *et al.*, 2005). Extrapolating these findings to the relationship between productivity (fertility) and species number may explain the recommendation of monocultures or bi-species mixtures for intensive agronomic systems. From our results and the discussion above several points emerge: (i) there may be differences between nutrient-rich and nutrient-poor ecosystems in which plant species, ecosystem processes and plant-to-plant interactions play important roles; (ii) results on diversity effects recorded in nutrient-poor ecosystems can, thus, not be extrapolated to nutrient-rich agroecosystems; (iii) even in fertile agroecosystems the highest yielding monoculture seems not to be the system with the highest performance.

It was not unexpected to observe at the Swiss site that increased nitrogen fertilization reduced the positive diversity effect. However, it was surprising that overyielding still persisted at the extremely high N levels of 450 kg N ha<sup>-1</sup> yr<sup>-1</sup>. It seems, therefore, that the mixtures tested also have an advantage under very intensive conditions. Under these non-limiting N conditions, the diversity effect cannot be explained by the symbiotic N<sub>2</sub> fixation of the legume component alone, especially since both N fixation and N fertilizer efficiency were reduced by increased N fertilization (Nyfeler *et al.*, 2008). This implies that nutrient retention in the system is improved in more diverse plant communities under low fertility conditions. Our results demonstrate that large amounts of fertilizer N can be saved in intensive forage production systems by the use of well balanced grass-legume mixtures. In this way we can both reduce environmental costs (e.g. N leaching and soil compaction) and economic costs of production (e.g. extra N fertilizer, extra fuel costs).

In contrast to other biodiversity experiments, the simplex design of the COST 852 experiment allowed us to determine the significance of species' evenness for diversity effects. We found that high evenness is a key factor for strong diversity effects. Reduced diversity effects under very high N fertilization at the Swiss site were accompanied by a strong reduction of evenness as the proportion of legumes in the sward strongly declined, probably as a result of competition for light. White clover with its stoloniferous growth form is especially known to be a weak competitor for light (Schwank *et al.*, 1986; Faurie *et al.*, 1996). Such difficulties in maintaining well balanced mixtures (Guckert and Hay, 2001) may be an important reason for the preference of grass monocultures in agriculture. It is a challenge for research in agronomy to develop management practices that help establish and maintain well balanced mixtures that maximise diversity effects. The simplex design and the analyses based on regression models are perfect tools to develop optimized mixtures with the corresponding relative species' abundances.

The importance of diversity may be underestimated if one considers only one of several ecosystem services that are provided by a system (Hector and Bagchi, 2007). For example, one ecological process may be maximized when species 1 and 2 are represented in a community, whereas another ecological process may only be delivered when species 3 and 4 are represented. This is a strong argument for the use of mixtures especially in the context of the ongoing development of agriculture towards multifunctionality (Durand *et al.*, 2002; Bertscher, 2004), thus emphasizing the importance of research for the practical application of mixtures. An important implication for future research is that the true functional benefits of diversity may only be appreciated when multiple important ecosystem processes are simultaneously considered. In an agricultural context, this could include factors such as yield, decomposition, nutrient leaching, pollination services and resistance to weed invasion. Indeed, our results show clearly that increased diversity strongly reduced the invasion of

unsown species. This was clearly demonstrated by the fact that the diversity effect was much greater when it was analysed in terms of sown species only. In addition, there are indications that species diversity seems to have had positive effects across trophic levels by increasing the microbial diversity and arthropod variability in the soil. This may have implications for decomposition of organic matter in the soil and, thus, nutrient availability to plants.

Looking to the future, it seems clear that species mixtures can deliver benefits to forage yield that are of significant magnitude, temporally persistent and spatially persistent at a geographical scale. An emerging challenge that faces both agronomists and ecologists is: what are the specific biological mechanisms that underpin these benefits of diversity? There are several well-known mechanisms by which diversity effects may be achieved. These include variations among different species in root architecture and canopy architecture that allow improved capture of available resources by more species-rich communities (complementarity), as well as inter-specific interactions that result in reduced incidence and transmission of pests and diseases in mixtures (facilitation e.g. Zhu *et al.*, 2000). In general, mixtures would also be expected to cope better with environmental heterogeneity. This could range from small-scale heterogeneity in nutrient availability and soil conditions, to the ability to recover from severe weather or management conditions. Investigating and resolving the degree to which such mechanisms contribute to diversity effects promises to be an exciting challenge!

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## **Session 1A**

# **Enhanced plant and animal diversity in semi-natural grasslands**

# Effects of agri-environmental agreements on acridids and plant species richness in alfalfa crops

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## Abstract

The decrease in food resources for farmland birds in Deux-Sèvres (France) is related to the decrease in grassland cover in agricultural landscape, and also to intensive management of alfalfa grasslands. Grasshoppers are among the major invertebrate inhabitants in grasslands and are central resources in the trophic network. Previous studies have shown that grasshopper abundances in alfalfa crops were rather low in the study area (intensive farmland in the south of the city of Niort in Deux-Sèvres) compared with other grasslands. In this study grasshopper abundances in alfalfa crops were investigated in relation to plant community structure and grassland management. The effectiveness of agri-environmental agreements in enhancing grasshopper abundance was evaluated. Results suggested that an extensive management of alfalfa crops (no pesticides and no cutting from the end of May to the end of August) enhanced grasshopper abundance and plant species richness. Plant community structure in alfalfa (number of plant species, number of botanical families, Shannon index) was linked to grasshopper abundance.

Keywords: grasshopper, locust, Gomphocerinae, biodiversity, population dynamics, artificial grasslands

## Introduction

Herbivorous invertebrates, and particularly acridids, are a dominant component of biodiversity in terrestrial ecosystems (Baldi and Kisbenedeck, 1999). In addition to the direct effect of reducing standing crop (Legg and Lockwood, 2001), acridids can influence ecosystem processes (Zhong-Wei *et al.*, 2006). They also play a major trophic role, being prey for other invertebrates and many vertebrates. In Europe, agricultural intensification has led to a major decrease in grassland cover over the last 30 years, and this has been accompanied by a more intensive use of the remaining grasslands. At the same time, biodiversity associated with grassland habitats has experienced dramatic declines in conjunction with habitat degradation and/or scarcity of food resources. In a previous study we described how different grassland types contribute to grasshopper abundance in an agricultural landscape where farmland birds are, to varying extents, of conservation concern and dependent on arthropods for their diet; we also have shown that acridid densities are rather low in alfalfa crops (Badenhausser and Bretagnolle, 2005). In this paper, we investigated whether agri-environmental subsidies help to maintain biodiversity, in this case acridids. We studied the effect of agricultural management intensity on acridid abundances in alfalfa crops in relation to plant community structure and management.

## Materials and methods

Fieldwork was conducted in 2006 in an area of mixed farming in the Poitou-Charentes Region (western France), with recent agricultural intensification. Twenty-eight plots (0.52 ha to 11.56

ha) were randomly selected among 1- and 2-year-old alfalfa plots, ten with agri-environmental agreements ('AEA') and eighteen without ('no-AEA'). The main purpose of these agreements was to avoid agricultural interventions in the plot between 15 May and 31 August, but fertilizers, insecticides and herbicides were tolerated from 1 November to 1 April. Acridids were sampled in all plots, while botanical observations were carried out in twenty plots (5 'AEA' and 15 'no AEA'). Moreover, plot management (fertilizer, herbicide, insecticide, number of cuttings) was available for 10 plots (5 'AEA' plots and 5 'no AEA' plots). Acridids were sampled on a field basis by means of a cage sampler (1 m x 1 m basis) thrown at 15 random sites within each field. Sampling was carried out during two periods. Immatures were sampled from 10-12 July and adults from 7-10 August. Immature individuals were counted and assigned to one of three groups: *Calliptamus* spp., Gomphocerinae sub-family, and all other acridid species. Adults were collected for later identification in the laboratory. Botanical observations were carried out from 10 April to 15 May, i.e., before the first cutting. Thirty points per plot were sampled using a 0.25 m<sup>2</sup> circle. Presence and absence of different plant species was noted. For each plot we calculated the number of plant species, Shannon index and equitability index to characterize plant species diversity. Splus software version 6.2 was used for the analysis of variance of mean number of individuals m<sup>-2</sup> per plot, using a square root transformation or logarithm transformation, and for regression analysis between acridid abundances and plant community structure.

## Results and discussion

Densities of immature individuals ranged from 0.07 to 17.4 m<sup>-2</sup> while adult densities ranged from 0 to 5.66 m<sup>-2</sup>. Dominant adult species were *Calliptamus italicus* and *Chorthippus* gr. *biguttulus* and *Euchorthippus elegantulus*. Agri-environmental agreements had a significant positive effect on all acridid abundances during both periods (Table 1) and there was no crop-age effect on acridid abundance. Densities of Gomphocerinae in AEA plots were 3 to 4 times of the densities observed in no-AEA plots, while *C. italicus* was 4 to 13 times more abundant in plots with agri-environmental agreements.

Table 1. Acridid density in alfalfa plots with or without Agri-Environmental Agreements (AEA plots, no-AEA plots, respectively) and ANOVA results.

	Acridid density m <sup>-2</sup> (SE) in 'AEA' plots (n = 10)	Acridid density m <sup>-2</sup> (SE) in 'no AEA' plots (n = 18)	F(ANOVA)	d.f.	Pr(F)
Gomphocerinae					
juveniles	1.72 (0.30)	0.40 (0.13)	23.8	1;26	< 0.001
<i>C. italicus</i>					
juveniles	5.21 (1.64)	0.40 (0.09)	19.9	1;26	< 0.001
Total juveniles	7.05 (1.72)	0.85 (0.16)	32.1	1;26	< 0.001
Gomphocerinae					
adults	1.18 (0.27)	0.40 (0.15)	10.6	1;26	< 0.003
<i>C. italicus</i> adults	1.01 (0.28)	0.25 (0.07)	10.7	1;26	< 0.001
Total adults	2.29 (0.49)	0.69 (0.17)	14.9	1;26	< 0.001

The number of plant species in alfalfa plots ranged from 4 to 43, and differed between 'AEA' plots (mean ± SE = 31.8 ± 4.2) and 'no AEA' plots (mean ± SE = 17.0 ± 2.0). Acridid abundance during immature and adult stages was not statistically ( $\alpha = 0.10$ ) related to plant species number in 'AEA' plots (linear regression analysis for immatures and adults: Pr(F<sub>I</sub>) = 0.22 and Pr(F<sub>A</sub>) = 0.27, d.f. = 1;3), nor in 'no AEA' plots (Pr(F<sub>I</sub>) = 0.91 and Pr(F<sub>A</sub>) = 0.55; d.f. = 1;13). The main difference between fields with and without agri-environmental agreements was the second cutting in fields without agreements, which generally occurs at the end of July. Negative effects of cutting have been found for acridid species, for example *Chorthippus*

*parallelus* (Guido and Gianelle, 2001). However, 'AEA' scheme did not have constraints about insecticide or herbicide treatments from September to May. The detailed crop management plans which were available for 10 plots showed that 'AEA' was associated with no insecticide treatment and with fewer herbicide treatments, but also that some 'no AEA' plots were associated with no insecticide treatments, and with no herbicide treatments. Thus, it is difficult to conclude which factors caused enhanced acridid abundance and plant species richness. Direct and indirect effects can be associated and masked by each other. However, our results suggest that there was a positive relationship between acridid abundance and plant diversity (number of plant species, Shannon Index). This relationship was not statistically significant in 'AEA' plots due to the low number of plots. In 'no AEA' plots, insecticide and/or herbicide treatments were generally used, and they masked the relationship between acridid abundance and plant species richness. When considering plots which received no insecticide treatments (5 'AEA' plots and 2 'no AEA' plots), there was a positive linear relationship between acridid abundance (transformed in square root) and plant species number ( $\Pr(F_1) = 0.04$  and  $\Pr(F_A) = 0.08$  d.f. = 1;5). This is in line with Perner *et al.* (2005) who suggested that arthropod abundance and especially herbivores were correlated with plant diversity.

## Conclusions

Agri-environmental agreements that reduce the number of cuttings and which are associated with no insecticide treatments and with fewer herbicide treatments, contributed to enhance plant diversity and acridid abundance in alfalfa plots. A positive relationship was established between acridid abundance and plant species richness in alfalfa plots without insecticide treatments.

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# Farm structure had only marginal effects on grassland diversity at a plot and farm level

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## Abstract

Increases in farm size and production intensity (i.e. animal density) are measures taken towards more economic farming. The effects of farm structure on grassland diversity, however, are unknown. We examined the effect of farm structure on grassland diversity (i) at the plot (species richness) and (ii) the farm level (proportion of species-rich grassland), by studying 25 farms with different sizes (4.6 to 52.0 ha) and animal density (0.4 to 2.4 cattle equivalents per ha) in a factorial design. Generally the proportion of species-rich grassland did not depend on the farm structure and was about 30% of the agricultural area, except in farms of small size and low animal density where it was 66%. The examined species-rich grasslands contained up to 71 species (per 25 m<sup>2</sup>). While site factors (altitude, slope) and management intensity explained 14.8 and 14.9% of variation in species composition at the plot level, farm size and animal density had no effect. Plots with high species richness were located mainly on marginal sites on steep slopes. We conclude that for nature conservation it is more important to support the cultivation of marginal sites than specific farm structures.

Keywords: species rich grassland, farm size, animal density, farm structure, botanical composition, Swiss Alps

## Introduction

Permanent grassland is widely distributed in the Swiss Alps. It is important for agricultural production and valuable as a habitat for numerous rare organisms. In the future, continuing structural changes in the agricultural sector will lead to increases in farm size and changes in production intensity (i.e. animal density). Since the effects of farm structure on grassland diversity are unknown, we examined the effects of farm size and animal density on two aspects of grassland diversity: (i) species richness at the plot level, and (ii) proportion of species-rich grassland at the farm level. Since farms with a high animal density need more high quality forage and produce a lot of farm manure we expected lower grassland diversity in terms of (i) lower species richness per plot and (ii) lower proportion of species-rich grassland on their agricultural area.

## Materials and methods

*Species composition at plot level:* In an alpine region in Grisons, eastern Switzerland, we selected 25 farms for a factorial design with five farm types (5 farms per type = 5 replicates) varying the two factors farm size (4.6 to 52.0 ha) and animal density (0.4 to 2.4 cattle equivalents per ha). On each farm four plots were selected according to management intensity and altitude (extensively and intensively managed each at low and high altitude). On each of these 97 plots (25 farms x 4 plots, 3 missing values) the yield fraction of all species was recorded on an area of 25 m<sup>2</sup>. Data were statistically analysed with canonical correspondence

analysis (CCA) and the Monte Carlo permutation test (CANOCO, Ter Braak and Šmilauer, 2002).

*Proportion of species-rich grassland at farm level:* On a subset of the same farms (4 replicates per farm type) we mapped the vegetation of the whole agricultural area (in total 506 ha and 1033 plots). The vegetation was classified into units combining phytosociological groups with site factors according to Dietl *et al.* (1981). These vegetation units were strongly related to site factors, management intensity and species richness.

## Results and discussion

*Species composition at plot level:* In the ordination (Figure 1A) the botanical relevés grouped well according to management intensity and altitude. The first axis ( $\lambda_1 = 0.543$ ) represented the nutrient gradient and was correlated with the factors altitude, slope and species richness (Figure 1A). The intensively managed meadows below 1500 m asl showed small differences in the botanical composition among individual plots and thus cover only a small area in the ordination. This group had an average of 31 species (per 25 m<sup>2</sup>). In contrast, the extensively managed meadows were more species rich on both altitudes and showed greater differences in species composition between individual plots. On average, 45 species were found below 1500 m asl and 57 species (maximum 71) above 1500 m asl. These extensively managed species-rich meadows were mainly found on steep slopes and shallow soils (marginal sites). Canonical correspondence analysis (CCA) revealed that site conditions (altitude, slope) and management intensity had highly significant effects on species composition (CCA,  $P < 0.0002$ ) and explained 14.8% and 14.9% variation in species composition. In contrast, farm size and animal density had no effect (CCA,  $P > 0.05$ ) on species composition as visualized in the ordination (Figure 1B).

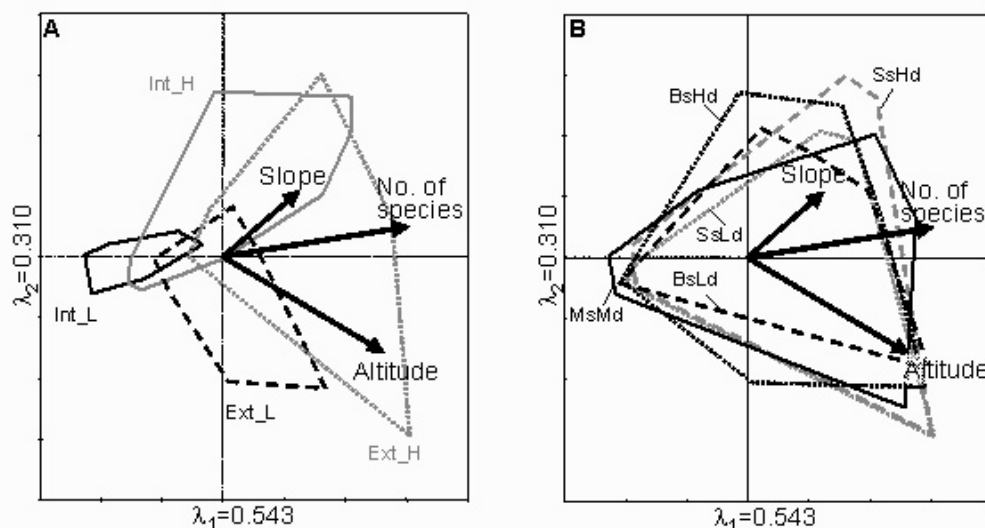


Figure 1. Variation in the vegetation of sites in a case study in the Swiss Alps revealed by correspondence analysis (CA) based on 97 relevés, grouped according to management intensity and altitude (A) and according to farm types (B). The envelope for each group is shown. The factors altitude, slope, number of species were passively added. A: Int and Ext = Intensively and Extensively managed; L and H = Low and High altitude. B: Bs, Ms and Ss = Big, Medium and Small farm size; Hd, Md and Ld = High, Medium and Low animal density.

These results demonstrate the dominant effect of site conditions and management intensity on species richness at the plot level. The reason that farm type had no effect on the species composition was due to (i) all farms having extensively managed plots and (ii) these plots

being managed under a comparable management regime on all farms. Two rules of the Swiss agri-environmental scheme played an important role: firstly, all farms must have a minimum of 7% of their farmland as an ecological compensation area (ECA) and secondly, all ECAs have to fulfil the same management restrictions (late cutting, no mineral fertilizer).

*Proportion of species-rich grassland:* Typical vegetation units of species-poor grassland were *Trifolium-Alopecuretum* or *Alchemilla-Cynosuretum*. These intensively managed ‘species-poor’ grasslands showed an average species number of 31, a high value when compared to species numbers in many lowland areas, e.g. 18 species in Germany (Hundt, 1983), 15 in the Netherlands (Kleijn *et al.*, 2001), 18 in England (Smart *et al.*, 2003). Species-rich grassland was represented by nutrient-poor meadows or pastures of the *Seslerietum*, *Nardion*, *Molinion* vegetation units. Their proportion of the agricultural area varied between 25 and 36% for all farm types except for small farms with low animal density (SsLd) where the proportion was 66% (Figure 2). Small farms with low animal density (SsLd) represented part-time or hobby farming, mostly with sheep. The farmer does not have to maximize his return and the available labour input is limited.

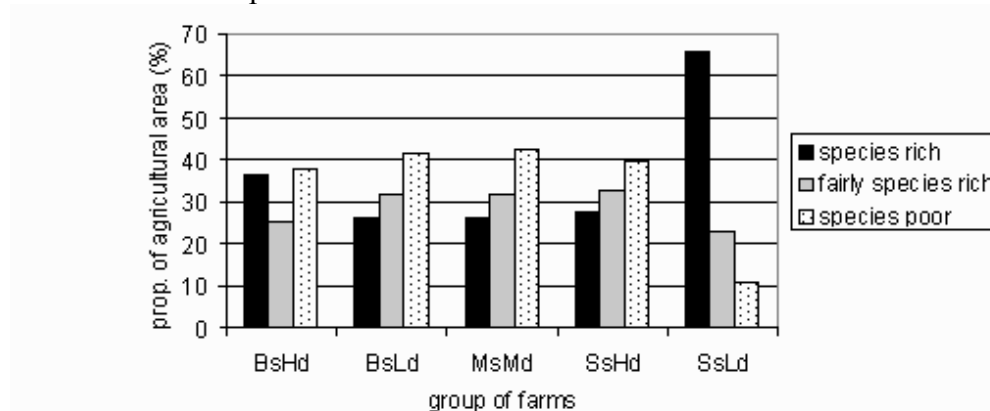


Figure 2. Effect of farm types on the proportion of grassland types on agricultural area. Bs, Ms and Ss = Big, Medium and Small size; Hd, Md and Ld = High, Medium and Low animal density.

## Conclusions

The farm structure (farm size and animal density) had no effect on the species composition and only minor effects on the proportion of species-rich grassland. However, species-rich grassland was found on marginal sites (steep slopes and shallow soil). Thus, for nature conservation it is more important to support the cultivation of marginal sites than specific farm structures.

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# Ecological rotation favours biodiversity in beef cattle systems in the French Massif Central

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## Abstract

In order to combine biodiversity preservation and production objectives in livestock farming systems, an 'ecological rotation' was developed, which required taking off the animals from some plots during the main flowering period, i.e. early June to early August. A 2-yr experiment investigated its feasibility and impact on vegetation and butterfly diversity. This rotation was compared under a rotational grazing system (RG) with continuous grazing (CG) at the same relatively high stocking rate (1.4 LU ha<sup>-1</sup>). Measurements were made on biomass, sward height, flowering intensity, and plant and butterfly diversity. In RG plots, butterfly species richness was 2.5 times higher and butterfly abundance twice that in the CG plots. Botanical diversity was not affected by grazing management, but flowering intensity was higher in the RG plots. This ecological rotation was carried out according to the initially planned schedule during a year with sufficient grass growth, but in an unfavourable year it reduced by 0.2 the number of livestock grazing days compared with the CG system.

Keywords: cattle, grazing management, semi-natural pastures, biodiversity, *Lepidoptera*

## Introduction

An empirical recommendation for increasing biodiversity in semi-natural grasslands is to adapt grazing periods, especially by taking off animals from some of the plots during the main flowering period. This allows plant species to achieve their reproductive cycle and offers food and shelter to insects, especially to nectar-feeding butterflies. As a consequence, farmers have to graze their animals at a higher stocking rate on the other plots, before grazing them on old reproductive swards in late summer on the plots previously managed for biodiversity. Finding an optimal balance between the preservation of biodiversity and the satisfaction of farmers' production objectives was one of the aims of this pilot experiment. A second aim was to evaluate the effect of taking off the animals from some of the plots during the flowering period on plant and butterfly diversity. The impact of this 'ecological rotation' was therefore compared in two successive years with that of a continuous grazing at the same relatively high stocking rate.

## Materials and Methods

The experiment was conducted in a semi-natural mountain pasture in the upland area of central France in 2005 and 2006. The study site had always been extensively managed, resulting in a high botanical diversity. In this pasture, two grazing treatments were set-up at the same relatively high stocking rate of 1.4 LU ha<sup>-1</sup> (1 LU = 600 kg liveweight): continuous grazing (CG) vs. ecological rotation (RG). Each management treatment was repeated three times according to a randomized block design in 3.6-ha plots. The RG plots were divided into four sub-plots (A, B, C and D), in which sub-plot D was taken out of the rotation during the main flowering period between early June and early August. Sub-plot D was grazed for 20 days in August (instead of 10 days for the other sub-plots), and then again for 10 days at the end of the grazing season, so that the total number of grazing days was similar in each sub-

plot. Seven 18-month-old heifers of Charolais and Limousin breeds were used in each block of each grazing management treatment. Heifers were turned out at pasture at the end of May until mid-October. Each year, sward height was measured at monthly intervals at 500 random locations per plot, and vegetation growth stage (vegetative, reproductive or dead material) was recorded at the first contact of a stick with the undisturbed sward surface. Herbage biomass was determined every month by cutting 5 m x 10 cm strips (0.5 m<sup>2</sup>) in the experimental plot. Five strips were cut in each CG plot, and 3 strips per sub-plot in each RG plot. One sub-sample per plot or sub-plot was taken to analyse grass nitrogen content in June, July and October. To quantify the botanical diversity, percentage cover of all plant species was estimated visually in ten fixed 1 m<sup>2</sup> quadrats per plot in July. The local abundance of nectar plants was also visually estimated in virtual 30 x 30 m<sup>2</sup> squares covering the whole plots by estimating the percentage cover of flowers. Butterflies were counted along fixed 50 m long by 5 m wide transects, with three transects per plot for CG and one transect per sub-plot for RG. All the butterflies encountered on a transect were counted and identified. Three counting periods were carried out between late June and early August. Faecal nitrogen concentration was used as an overall indicator of diet digestibility. The mixed ANOVA model included a fixed management treatment factor at the plot or sub-plot level, period, a random block factor, and the interactions between treatment and period, and treatment and block.

## Results and Discussion

In spite of annual variations ( $P < 0.01$ ), the species richness and abundance of butterflies were higher ( $P < 0.05$  and  $P < 0.001$ , respectively) in the ecological rotation compared with continuously grazed plots at the same relatively high stocking rate. The abundance and species richness of butterflies were positively related to flowering intensity ( $r = 0.74$ ,  $P < 0.01$ ;  $r = 0.73$ ,  $P < 0.01$ , respectively), and negatively correlated to the proportion of sward cover that was lower than 10 cm ( $r = -0.36$ ,  $P < 0.05$ ;  $r = -0.38$ ,  $P < 0.05$ ). Botanical diversity was not affected by plot management, but in each year flowering intensity was greater in the RG plots ( $P < 0.01$ ; Table 1).

Table 1: Butterfly, botanical diversity, and sward characteristics in the two treatments CG: continuous grazing vs. RG: ecological rotation (only the measurements taken in July are reported). For RG, italics indicate results from sub-plots ABC vs. sub-plot D.

	CG '05	RG'05	CG'06	RG'06	Treat. (T)	Year (Y)	T.*Y.
Butterfly species/trans.	1,3	3,4 (2,4-6,3)	1,6	3,8 (3,4-5,0)	***	***	*
Butterfly abundance/trans.	3,2	7,8 (4,1-19,0)	3,1	5,3 (3,6-10,7)	**	**	*
Plant species per plot	54	57	58	54	ns	ns	ns
Flowering intensity	0.5	1.0 (0,5-2,5)	0.8	1.3 (1,1-2,1)	**	**	ns
Biomasse (t DM ha <sup>-1</sup> )	2.9	3.0 (2.8-3.6)	1.8	0.8 (0.7-1.1)	t	***	*
Sward height (cm)	9.5	12.2 (10,7-16,8)	8.2	8.7 (6,7-14,8)	t	*	ns
Herbage N (g kg <sup>-1</sup> DM)	17.7	18.0 (18.1-17.6)	19.4	19.6 (19.3-20.2)	ns	*	ns

\*\*\* :  $P < 0.001$ ; \*\* :  $P < 0.01$ ; \* :  $P < 0.05$ ; t :  $P < 0.10$ .

In July 2005, average sward height was higher in the ecological rotation compared with continuously grazed plots, while herbage biomass and grazing intensity were similar (Table 1). Conversely, a similar sward height was measured in the two treatments in 2006, which was lower than that measured in 2005 as the result of weather conditions detrimental to grass growth in spring 2006. Herbage biomass was also lower in RG compared with CG. In order to have the sub-plot D ungrazed until early August, we had to take off two heifers out of each RG plot at the end of June, and then all the animals for one week in late July, whereas biomass production was sufficient in CG plots. RG and CG plots were thus managed according to the pre-planned grazing schedule in 2005, with a total of 580 LU-grazing days

during the grazing season. In 2006, under unfavourable grass growth conditions, grazing schedule had to be adapted and the number of LU-grazing days was 20% lower in RG compared with CG plots (418 vs. 529). Nitrogen content of herbage and faeces was never affected by the grazing management treatments. Clear differences were also observed between the different sub-plots in the RG system: species richness and abundance of butterflies were significantly higher in sub-plot D compared with sub-plots A, B and C from mid-July ( $P < 0.01$ ; Figure 1).

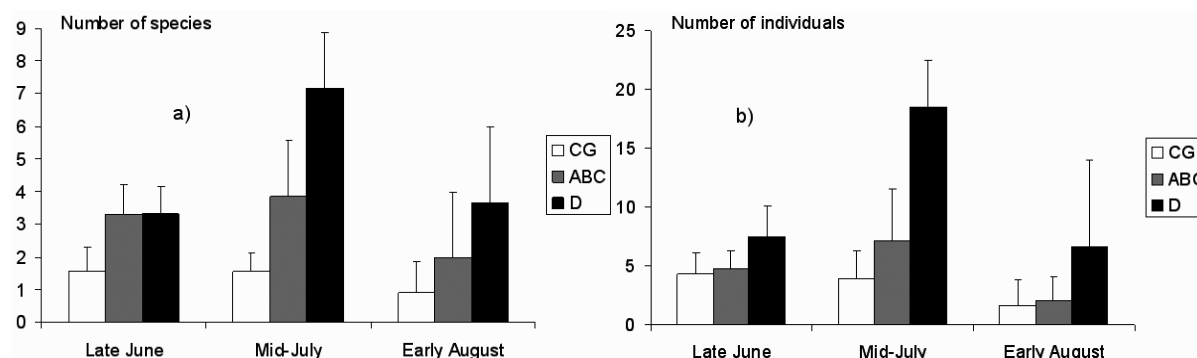


Figure 1. Seasonal variations of butterfly population per transect: a) Species richness, b) Butterfly abundance. Different shadings are for treatments, i.e. open for CG, grey for ABC sub-plots in RG, and black for sub-plot D.

In July, the species richness and abundance of butterflies were also higher in sub-plots ABC of RG compared with the CG plots (Table 1). This could be the result of the attractiveness of sub-plot D. Flowering intensity ( $P < 0.001$ ), herbage biomass ( $P < 0.10$ ) and sward height of sub-plot D ( $P < 0.05$ ) were also higher than that of sub-plots ABC (Table 1). The ecological rotation thus led to the creation of contrasting sward structures in the different sub-plots and enhanced flowering intensity. This had a positive impact on butterfly populations: in RG plots butterfly species richness was 2.5 times higher and butterfly abundance twice higher than in CG plots. This result confirms the positive impact of increasing the proportion of flowering plants in semi-natural pastures on butterfly populations (Öckinger *et al.*, 2006; Dumont *et al.*, 2008). However, the effects of this particular type of management need to be assessed in the long-term. Sub-plot D, having been attractive for adult butterflies during the flowering period, may act as a ‘trap plot’ for butterfly eggs and larvae when grazed again in August.

## Conclusion

This ecological rotation favoured butterfly populations in this diverse, semi-natural mountain pasture. Further observations are, however, needed to evaluate the long-term plant response, to test for the ‘trap-plot’ hypothesis, and to define the optimal stocking rate for such a management practice. Indeed, in the less favourable year for biomass production it revealed its potential cost in term of LU-grazing days. Its interest in fertile and less diverse pastures would also have to be tested.

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# Effects of plant litter on seedling establishment in semi-natural grasslands

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## Abstract

This study examines the impact of plant litter on seedling establishment in a semi-natural grassland. In a field experiment, seeds of six forbs were sown in plots subjected to either different litter quantities or addition of water extracts of litter. The response to plant litter was species specific. High quantities of litter resulted in a negative response in all species studied, except *Anthriscus sylvestris*. In some of the species, the response shifted from positive or neutral at intermediate litter quantities, to negative at high litter quantities. This suggests that facilitative effects were present, although inhibitory effects dominated at high litter quantities. The physical effects were generally stronger than the chemical effects. However, water extract of litter inhibited emergence in three of the six species. Filtration using activated carbon removed the negative effect of litter extract, which suggests that the effect was caused by inhibitory compounds adsorbed by activated carbon (e.g. polyphenols) rather than by increased competition in response to nutrients added via the extract.

Keywords: facilitation, germination, plant litter, seedling establishment, semi-natural grassland

## Introduction

Plant litter is an important determinant of vegetation dynamics and community structure due to its effects on germination and seedling growth. Semi-natural grasslands depend on regular management like mowing and grazing and when management ceases, the result is often accumulation of plant litter. The effect of litter is complex and depends on the balance between facilitative and inhibitory effects that occur simultaneously. Litter serves as a physical barrier, inhibits the transition of light, reduces the diurnal fluctuations in temperature and alleviates the moisture stress. Litter also interacts with invertebrates, micro-organisms and fungi that influence the survival of seeds and seedlings. Compounds released from litter influence competitive relationships among plants, and between plants and soil microbes (Fenner and Thompson, 2005). Secondary plant metabolites, e.g. polyphenols, released from litter can have direct inhibitory effects, whereas nutrients can trigger germination and seedling growth (Fenner and Thompson, 2005). This study focused on the effect of plant litter on seedling establishment in six forb species in semi-natural grasslands.

## Materials and methods

A sowing experiment was conducted in a semi-natural grassland at the Norwegian University of Life Sciences, south-eastern Norway. Seeds were sown in September 2003 and seedling emergence and establishment were recorded in 2004 (total precipitation was 841 mm and mean temperature 6.5°C in 2004). The soil was well drained and nutrient-poor and the vegetation short and dominated by grasses. The experiment included ten blocks, each with six plots of 0.6 x 0.6 m randomly assigned to six different treatments. In the centre of each treatment plot, seeds of the six species were sown in plots of 0.4 x 0.4 m. The six species (Figure 1) are forbs common to semi-natural grasslands in southern Norway. A description of the species is given in Hovstad and Ohlson (2008). The experimental treatments included



(1) a control without litter, (2) 400 g litter m<sup>-2</sup> and (3) 900 g litter m<sup>-2</sup>. The effect of compounds leached from the litter was estimated using two water extracts: (4) low and (5) high concentration corresponding to 400 and 900 g litter m<sup>-2</sup>. To estimate the effect of secondary chemical compounds, the experiment included (6) addition of extract treated with activated carbon. Polyphenols and other secondary plant metabolites are adsorbed by activated carbon and can be removed by using a filter (Hille and den Ouden, 2005). Filtration had no significant effect on the concentration of nitrogen, carbohydrates and total organic carbon in the extract (Hovstad and Ohlson, 2008). Plots assigned to extract treatments were given one litre of the respective extract each week in a six-week period from April 28 to June 2. Plots without addition of extract were treated with a corresponding amount of water.

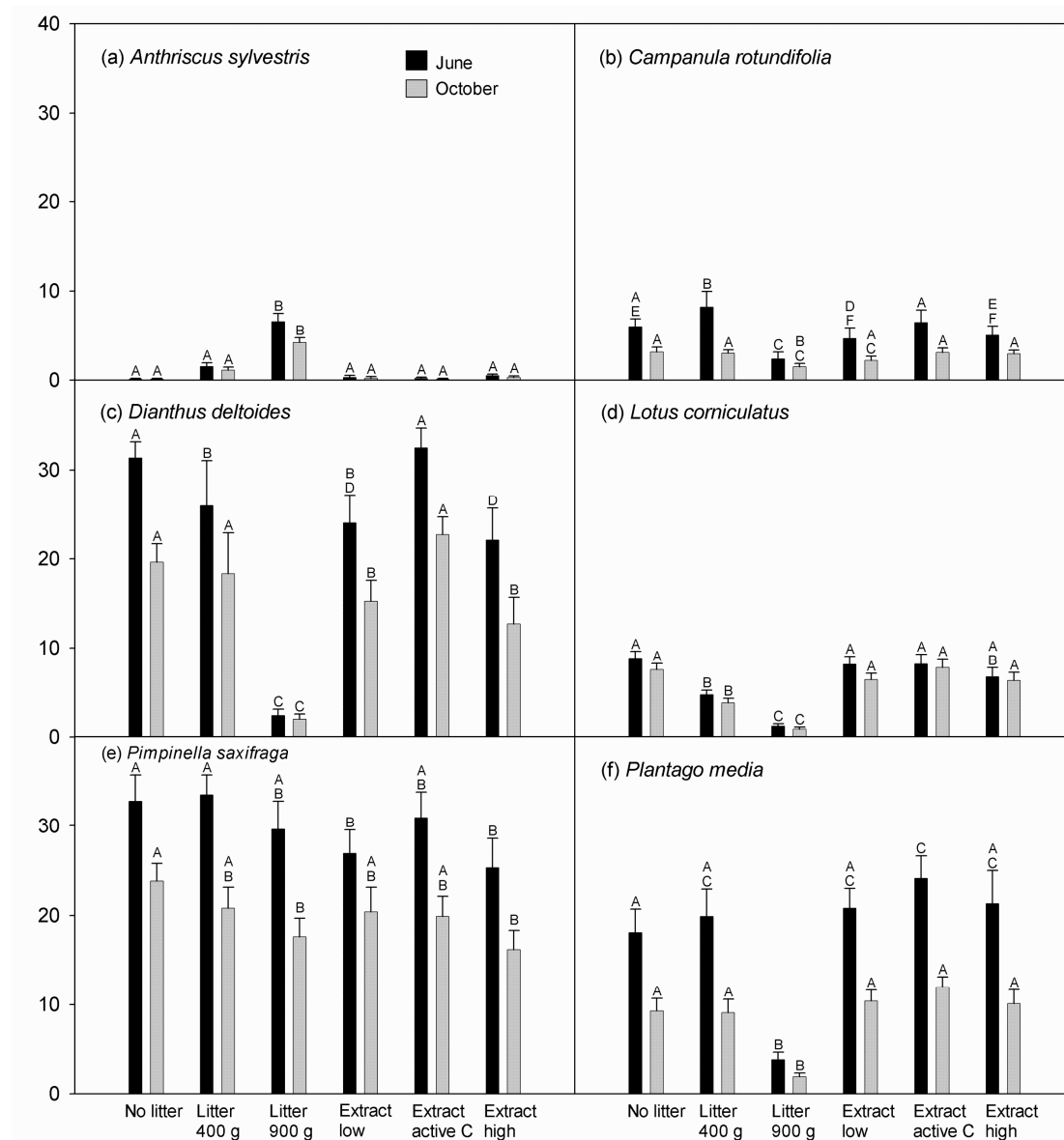


Figure 1. Seedlings per 100 seeds (mean  $\pm$  SE) at the census in June (black bars) and October (grey bars) in plots with different treatments. Within each census, different letters represent significant differences.

A detailed description of design and procedures is given in Hovstad and Ohlson (2008). Seedlings were counted in the periods June 22-27 and October 12-18, and the vegetation was not mown until after the last census. Seedling emergence and establishment, i.e. the

proportion of seedlings per 100 seeds, were assumed to have a binomial distribution and analysed using generalized linear models. Correlations between seedling numbers of different species within the same plot were accounted for using a mixed modelling approach.

## Results and discussion

The response to plant litter depended on both species identity and litter quantity (Figure 1). The three species *Dianthus deltoides*, *Pimpinella saxifraga* and *Plantago media* displayed no response, or only a minor negative response, to 400 g litter m<sup>-2</sup> but a clear negative response to 900 g litter m<sup>-2</sup>. In contrast, *Lotus corniculatus* had a clear negative response to both levels of litter proportional to litter quantity. In *L. corniculatus*, germination is promoted by a period of low and fluctuating temperatures (Van Assche *et al.*, 2003). In this experiment, litter reduced the diurnal fluctuations in temperature at the soil surface (see Hovstad and Ohlson, 2008) and this may have contributed to the negative effect of litter. In *Campanula rotundifolia*, intermediate quantities of litter had a positive effect on seedling emergence, i.e. facilitative effects were more important than inhibitory effects. Possible facilitative effects are reduced water stress or a positive interaction between litter and mycorrhiza, which increases growth and vigour of *C. rotundifolia* seedlings (Grime *et al.*, 1988). Germination in *C. rotundifolia* is promoted by light (Grime *et al.*, 1988) and high quantities of litter had a negative effect. *Anthriscus sylvestris* is the only species with a positive response to high quantities of litter and often increases in abundance when management ceases. It has been suggested that some species, or functional groups, are facilitated by their own litter and that this contribute to successional shifts in the vegetation (Quested and Eriksson, 2006). However, experiments have shown that *A. sylvestris* is not facilitated by its own litter any more than by litter from other species (Hovstad, 2007). The results displayed no consistent relationship between seed mass and response to plant litter. Litter had a large effect on seedling numbers compared with the effect of water extract of litter. However, the litter extract inhibited seedling emergence in three of the six species (Figure 1). The litter extract treated with activated carbon had no negative effect on seedling numbers. This indicates that the inhibitory effect was caused by compounds that could be adsorbed by activated carbon.

## Conclusions

The response to plant litter varied among species. Positive, neutral and negative responses were observed at intermediate litter quantities, whereas negative responses dominated at high quantities. Although the effects of litter were mostly physical, the results indicate that inhibitory compounds leached from litter can affect seedling numbers in some species.

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# Troublesome plants in species-rich grasslands: can we maintain the toxic plant *Carex brevicollis* and alleviate its risk to livestock?

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## Abstract

*Carex brevicollis* (DC in Lam. & DC) is a sedge naturally distributed on limestone, species-rich grasslands in the central and southern mountains of Europe. In areas where the species is abundant, frequent abortions associated with its consumption occur in pregnant sheep, cows and mares. Published studies indicate a high content of toxic alkaloids in *C. brevicollis* leaves. The toxicosis has led some farmers to the abandonment of grazing areas and, in some cases, to the controversial demand of measures for removing the species from grasslands. In order to ensure its maintenance and alleviate the risk to livestock, this research focused on 1) the nature of the alkaloids, and 2) the grazing practices followed by farmers. Toxic alkaloids seemed to be produced by the plant, but their concentrations were highly enhanced by the occurrence of endophyte fungal infection. This result highlights the complexity of the interactions plant-fungus-mammals and encourages further research. Independently, some farm management strategies may be implemented to decrease the risk of abortions, including the avoidance of grazing frost-covered pastures, the supplementation at starvation periods and the maintenance of the offspring with the herd during the summer grazing period.

Keywords: livestock toxicity, *Carex brevicollis*, grazing practices, endophytes, alkaloids

## Introduction

Urbasa-Andia Natural Park is a large, mountain area located south of the western Pyrenees (42°48'-42°52'N and 1°22'-1°32'E; average elevation 950m a.s.l.), occupied by grasslands, heathlands and beech forests of great floristic richness. It comprises two limestone ranges: Urbasa (11,400 ha) and Andia (4,700 ha). The use of these rangelands is regulated by an ancestral right which allows the free-range of any domestic flock of the Navarra county. Because of this, livestock movements have occurred since historical times. Nowadays, 34,700 sheep, 4,200 cattle and 2,600 horses belonging to 310 farmers graze extensively the area during the spring, summer and autumn period.

In 2006, we undertook the Grazing Management Planning of the Park. One concern was the knowledge of the main risks associated to the extensive grazing. Farmers' opinions are summarized in Figure 1. Risks were similar between ranges, with the exception of poisonous plants, that mostly bothered Urbasa's farmers. The toxicity was mainly associated with the consumption of the sedge *Carex brevicollis* (DC in Lam. & DC), which grows in Urbasa but not in Andia. This sedge is a common component of two protected natural habitats (directive 92/43/EEC): limestone subalpine grasslands and calcicolous beech forests. Previous published studies indicate a high content of the alkaloid brevicolline in the leaves of this species (Sharipov *et al.*, 1975). This alkaloid has an intense oxytocic effect, enhancing uterine contractions and causing abortions in pregnant mammals (Marcu, 1965). Despite the risk, no literature has been found on the origin of the alkaloid (intrinsic or extrinsic to the plant), nor about the implementation of grazing strategies to avoid or minimize poisoning. In order to gain insight about it, we undertook a multi-faceted research focused on two main issues: 1) the origin of the toxin, and 2) the grazing practices followed by farmers. Regarding the first

point, we were interested in knowing whether the alkaloids were produced by the plant or by an associated endophytic fungus. In the second case, measures to reduce the percentage of fungal infection in the natural populations could potentially be implemented.

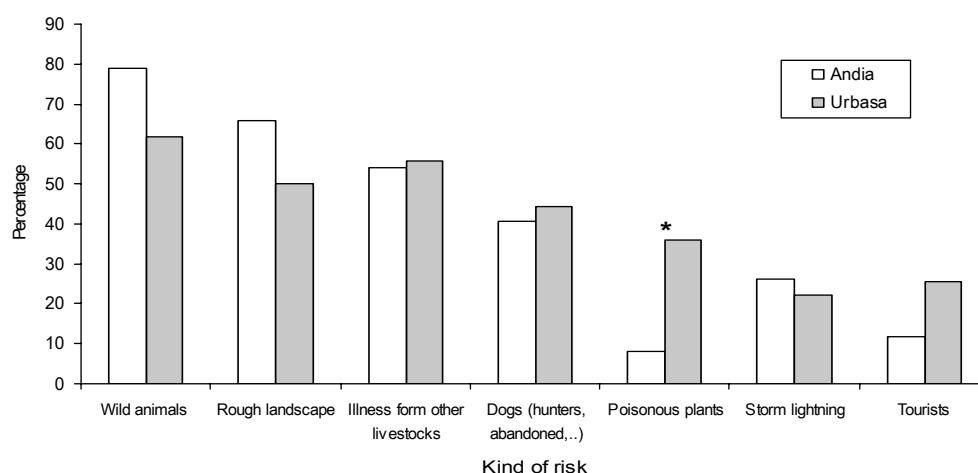


Figure 1. Main risks associated to grazing in the Park. Andia farmers worried about poisonous plants are horse owners, who move freely from Andia to Urbasa rangelands (\* significant at  $P > 0.05$ ).

## Methods

*Investigation of the origin of the toxin.* In autumn 2006 we collected 12 plants of *C. brevicollis* from three Urbasa grassland areas (Udau, Bardoitza and Tximista). A leaf tissue sample of each plant was obtained and analysed by microscopy for the occurrence of endophyte infection (aniline blue method of Latch *et al.* (1988)). We found associated fungal endophytes in nearly 40% of the samples. A pot experiment was therefore set up in which *C. brevicollis* plants were planted in separate pots and maintained in a greenhouse. After several weeks of growth, four tillers per pot were transplanted. One tiller was kept as a control and the remaining three were fumigated with the fungicide Propiconazole (800µg/treatment, 3 times at 10-day intervals) in order to kill the fungi. The fungal treatment was effective since the infection rate was reduced to 8% of samples. After a new period of growth, newly grown tillers were collected, and alkaloids were extracted (Zayed and Wink, 2005) and analysed quantitatively for brevicolline by GLC-MS.

*Survey of farmers.* During the summer/autumn 2006 we undertook an extensive survey on Urbasa farmers, in order to determine their livestock toxicity problems, the grazing areas frequented and the extensive management practices followed. Ninety-one farmers were surveyed; they owned 11,526 sheep, 2,074 cows and 560 mares.

## Results and discussion

The alkaloid brevicolline seemed to be produced by the plant, since all individuals, irrespective of the treatment, displayed some content (Table 1). However, concentrations were significantly much lower in fumigated, fungal-free, plants ( $F = 7.522$ ,  $P = 0.029$ ), which indicated that the presence of fungus favoured, to some extent, the synthesis of the alkaloid. These results require further investigation and will be the aim of future research. We are particularly interested in determining which fungus, or group of fungi, enhances the alkaloid synthesis, the mechanistic explanation of the process and the ecological consequences of it.

Table 1. GLC-MS results of the brevicolline analyses.

	N. molecules of brevicolline	
	Mean	Std error
Mother plants	1,672,438.5a	445,526.7
Non-fumigated tillers	1,079,741.2a	251,360.4
Fumigated tillers	188,991.4b	50,322.8

Table 2 summarizes the results of the farmers' survey. The consumption of *C. brevicollis* caused abortions in the three livestock species present in the Park, cows, sheep and mares, but mostly in young pregnant animals. Management approaches followed to avoid the poisoning varied between livestock species. Among sheep farmers, strategies implemented to avoid toxicity included: 1) maintenance of young ewes with the flock during the first highland grazing period, in order to nurse with the mother and get introduced to its food preferences; 2) fodder supplementation to avoid starvation, and 3) abandonment of the grassland before first frosts, which cover *C. brevicollis* and make its detection difficult. In the case of cows, farmers indicated that the practice of maintaining young calves with their mothers during the highland period conflicted with other interests (production of one calf per year, highly demanding needs of pregnant cows better ensured without nursing, more difficulty of calf breeding in low-productive pasturelands). According to this, forage supplementation and the early abandoning of the area were the strategies most considered. For horses, in contrast to other livestock, offspring usually stay with the mare during the highland grazing period. However, this traineeship does not appear to build up a selective skill and to prevent toxicities in future mares. In fact, only farmers that left or moved to other grazing areas prevented poisoning. Some horse farmers have demanded a permanent fence between the two ranges in order to avoid the entry of the animals to Urbasa rangelands.

Table 2. Urbasa farmers survey results. Risk of poisoning and strategies implemented.

	Sheep farmers	Cattle farmers	Horse farmers
Total number of farmers in the Urbasa rangeland	36	39	16
N. of farmers with livestock in <i>Carex brevicollis</i> areas	19	23	7
Farmers not affected by poisonings	7	16	3
Not affected farmer's practices <sup>(1)</sup> :			
Mothers and offspring graze together	3	0	<i>usual practice</i>
Leave the area or move before first frosts	2	8	3
Supplement when grass is scarce	3	9	1
Graze in an area of low stocking rate	1	0	0

(1) More than one practice may be done

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# **Vegetation and butterfly diversity in a species-rich mountain pasture grazed by cattle under different stocking rates**

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## **Abstract**

The relationships between vegetation and butterfly diversity were analysed in a species-rich mountain pasture of central France, continuously grazed by heifers under three different stocking rates: 0.6, 1.0 and 1.4 livestock units per hectare. At the plot scale, the evolution of forbs and butterflies followed the same pattern, the abundance of both groups increasing in plots grazed at a low stocking rate and over the 5-yr experimental period. Species richness of butterflies also increased at the two lowest stocking rates ( $P < 0.001$ ), and rose from 7 to 15 species per plot ( $P < 0.001$ ) over the duration of the experiment. Detailed observations in 30 x 30 m<sup>2</sup> squares revealed a negative correlation between the abundance of nectar plants and grazing intensity. Numbers of butterflies that were counted along 50m long by 5 m wide transects also increased with the local abundance of nectar plants ( $r = 0.56$ ;  $P < 0.001$ ), and consistently decreased with local grazing intensity ( $r = -0.62$ ;  $P < 0.001$ ). Stocking rate thus had a great impact on butterfly populations through the alteration of vegetation structure and composition. Tall vegetation in plots grazed at a low stocking rate provided greater structural heterogeneity as well as more nectar resources for the butterflies.

Keywords: cattle grazing, stocking rate, nectar plants, Lepidoptera, semi-natural pastures

## **Introduction**

Maintaining biodiversity in semi-natural grasslands is one of the major challenges for biodiversity conservation in Europe. Diversity has frequently been assumed to follow a unimodal response along disturbance gradients (Huston, 1994). Though manipulation of stocking rate is probably the most commonly used management practice in grazed areas, there are few targeted experiments whose results support this hypothesis in order to make practical management applications on the basis of reliable data. Here, we opted for using vascular plants and butterflies, which have different ecological characteristics (Pöyry *et al.*, 2006), to test for more general and applicable patterns of diversity in relation to a reduction of stocking rate. We focused on the relationships between butterflies, sward structure and nectar plants in order to understand the effect of reducing stocking rate on butterfly diversity.

## **Materials and methods**

The study area was set up at an altitude of 1100 m in an open landscape of central France (45°15'N, 2°51'E) characterized by a high initial level of diversity. Upland areas are refuge areas for butterfly populations that suffer from a general decline as the consequence of intensification in lowland agricultural areas. In total, 51 different butterfly species were observed on the study site. The pasture itself included 170 different plant species, with a majority of oligotrophic species due to the low soil nutrient status, and this in spite of a rather intensive grazing history: 1.75 livestock units (LU) ha<sup>-1</sup> (with 1 LU = 600 kg liveweight)

between 1985 and 1991, reduced to 1.15 LU ha<sup>-1</sup> between 1992 and 2001. During five years (2002-06), we recorded the abundance and diversity of vascular plants and butterflies in 3.6-ha plots, continuously grazed by heifers at three different stocking rates: 0.6, 1.0 and 1.4 LU ha<sup>-1</sup>. Each stocking rate treatment was repeated three times according to a randomized block design. Measurements were made in July after differences in sward structure had been established between the stocking rate treatments (Dumont *et al.*, 2007). To evaluate the botanical diversity, percentage cover of all plant species was estimated in ten fixed 1 m<sup>2</sup> quadrats per plot. Any additional species occurring within a 5 x 5 m<sup>2</sup> area centred on each quadrat were recorded and assigned a conventional cover value of 0.1. Grazing intensity and the local abundance of nectar plants were also visually estimated based on a 30 x 30 m<sup>2</sup> squared grid covering the whole plots. Butterflies were counted on three occasions each summer along three fixed transects (50m long by 5 m wide) per plot. Data were averaged at the plot scale, but could also be correlated to the local sward structure and local abundance of nectar plants using the linear regression model of SAS and considering the six closer 30 x 30 m<sup>2</sup> squares along each transect. At the plot scale, in addition to a fixed stocking rate factor, the mixed ANOVA model included a year factor, a random block factor, and the interaction between stocking rate and year. We used the same model to analyse the evolution of plant species abundance, considering their initial abundance in 2002 as a covariate. Differences between treatments were detected using the Tukey-Kramer correction for multiple comparisons.

## Results and discussion

Over the 5-yr measurement period, a reduction of stocking rate from 1.4 to 0.6 LU ha<sup>-1</sup> did not affect the number of plant species per plot, but slightly reduced the number of plant species per m<sup>2</sup> (Table 1). Conversely, forb abundance increased from 29.9 to 34.8% of plot area between 2002 and 2006 ( $P < 0.01$ ), and was also greater in plots grazed at the lowest stocking rate ( $P < 0.01$ ). This was particularly true for tall species such as *Achillea millefolium*, and for species that can develop in tall vegetation patches such as *Helianthemum nummularium* and *Galium verum*. Measurements made in the 30 x 30m<sup>2</sup> squares confirmed a negative correlation between the abundance of nectar plants and grazing intensity ( $r = -0.40$ ;  $P < 0.001$ ).

Table 1. Response of botanical and butterfly diversity to the stocking rate (SR) gradient.

Botanical diversity	1.4 LU ha <sup>-1</sup>	1.0 LU ha <sup>-1</sup>	0.6 LU ha <sup>-1</sup>	s.e.	SR	Year (Y.)	SR x Y.
Species richness / paddock	56.0	55.1	53.0	3.2	ns	ns	ns
Species richness / m <sup>2</sup>	25.4a	25.1a	23.8b	1.1	**	**	ns
Forb abundance (%)	28.1a	30.9a	36.2b	2.2	**	**	ns
<i>Achillea m.</i> (%)	4.35a	3.86a	5.63b	0.54	**	***	ns
<i>Galium v.</i> (%)	2.21a	2.48a	6.95b	0.64	***	*	ns
<i>Helianthemum n.</i> (%)	1.65a	3.43ab	4.39b	1.08	*	ns	ns
Butterfly diversity							
Species richness / paddock	7.7a	12.5b	11.3b	0.7	***	***	0.06
Individuals / paddock	28.2a	43.3b	63.0c	3.8	***	***	0.09
- from tall vegetation	22.7a	32.2a	51.8b	3.3	***	***	ns
- of short grassland	2.9	5.1	4.6	0.8	0.06	**	ns
- dependent of forbs	5.8 a	15.1b	16.5b	2.2	***	***	*
<i>Thymelicus lineolus</i>	3.7a	9.7a	21.4b	2.1	***	***	*
<i>Maniola jurtina</i>	8.9ab	8.1a	12.3b	1.1	*	ns	ns

Means with different superscripts differ; \*\*\* :  $P < 0.001$ ; \*\* :  $P < 0.01$ ; \* :  $P < 0.05$ .

At the plot scale, the response of butterflies to a reduction in the stocking rate was similar to that of forb species (Table 1). The abundance and species richness of butterflies increased

from an average of 28 to 67 butterflies per plot ( $P < 0.001$ ), and from 7 to 15 species per plot ( $P < 0.001$ ) between 2002 and 2006. This increase tended to be stronger in plots grazed at a low stocking rate (stocking rate  $\times$  year:  $P = 0.09$  for abundance and  $P = 0.06$  for species richness). The number of butterflies from species associated with tall vegetation (including *Thymelicus lineolus* and *Maniola jurtina*) increased in plots grazed at a low stocking rate, as well as those from species whose larval stage depends on forbs (Table 1). The interaction between stocking rate and year was significant for these forb-dependent species ( $P < 0.05$ ), with a four-times increase in their population between 2002 and 2006 in plots grazed at the two lowest stocking rates, compared with only a doubling of population size at 1.4 LU ha<sup>-1</sup>. Interestingly, the positive effect of a lenient grazing intensity was also found for butterfly species associated with short grasslands ( $P = 0.06$ ), which confirms the benefit of increased structural heterogeneity through patchiness already discussed by Dennis (2004). Finally, butterfly abundance along the 50-m long by 5-m wide transects increased with the local abundance of nectar plants ( $r = 0.56$ ;  $P < 0.001$ ), and consistently decreased with local grazing intensity ( $r = -0.62$ ;  $P < 0.001$ ).

At both scales, stocking rate thus had a great impact on butterfly populations through the alteration of vegetation structure (Dumont *et al.*, 2007) and composition (Table 1). Tall vegetation in plots grazed at a low stocking rate provided a greater structural heterogeneity as well as more nectar resources for the butterflies. This is consistent with the observation made by Öckinger *et al.* (2006) that sheep grazing could be detrimental to butterfly populations as the result of their selectivity for nectar plants, and resulting impact on sward composition. Our results suggest that species richness of butterflies would, however, peak in taller vegetation compared with vascular plants, which confirms previous reports (Pöyry *et al.*, 2006).

## Conclusion

A reduction of stocking rate in this species-rich mountain pasture of central France led to more structural heterogeneity and feeding resources for butterflies, and consequently to an increase in their abundance and species richness. A key issue in grassland ecology is the enhancement of vegetation heterogeneity, so that policy framework and management practices that promote the diversity of farming systems and landscape heterogeneity have been seen as the key to restore and sustain biodiversity in livestock farming systems (Benton *et al.*, 2003). Consistently, the contrasting stocking rates applied to the different plots in the present study area, together with a slight decrease in average stocking rate from 1.15 to 1.0 LU ha<sup>-1</sup>, favoured butterfly diversity as indicated by the doubling of their population over the 5-yr measurement period.

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# Effect of fodder plantations of *Atriplex canescens* on the floristic diversity at nine stations in the department of Laghouat (Algeria)

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## Abstract

In the Algerian steppes overgrazing combined with dryness leads to degradation and loss of vegetation cover. To meet the growing needs of livestock, the government launched an operation to plant 1 million ha of *Atriplex canescens*. To determine the impact on floristic diversity of the plantation of this species a methodology was adopted based on the comparison of 27 phyto-ecological statements carried out inside and outside the plantations. This study shows there was a therophytisation of plantations with a quantitative and qualitative improvement in species. This is attributed to the role of *A. canescens* in conserving soil moisture and through trapping sand and thus allowing self-mulching by annuals. The systematic analysis shows an important diversity in families and an important generic and specific richness. The principal families represented were *Asteraceae*, *Poaceae* and *Fabaceae*. The analysis of the biological spectra shows the predominance of therophytes, species producing many seeds that colonize open spaces quickly.

Keywords: Algeria, steppe, biodiversity, plantations, *Atriplex canescens*

## Introduction

Increased needs for livestock fodder in Algeria during the past three decades has lead to the Algerian government adopting the plantation of fodder seedlings such as *Atriplex canescens*. Our study was carried out in nine plantations of *Atriplex canescens* characterized by different soils in Laghouat's steppes. This study was carried to observe the impact of this species on the floristic diversity. The nine stations are located in South Algiers' steppes; this zone is characterized by annual average precipitation of 240 mm and annual average temperature of 22°C The duration of the dry period is 7 months.

## Methodology

For this study, we adopted a methodological step based on the comparison of the phytoecological statements between the planted and non planted stations. The site of the 27 statements is dependent on the physiognomic and geomorphologic homogeneity of the stations. Using the method of Braun Blanquet, we determined a minimal surface of 64 m<sup>2</sup>, in which we counted the species present inside and outside the plantations. These were characterized from the biological point of view according to the classification of Raunkiaer amended by Ellenberg *et al.* (1967). To highlight the impact of the introduction of *Atriplex canescens*, we evaluated the differences in floristic changes between the inside and the outside of the plantations, and for each station we calculated:

$$\text{The Sorensen index } \frac{2c}{a+b} \times 100$$

The rate of pastoral species variation by pastoral class index: the species were classified according to their specific quality index (IS) (C.R.B.T, 1978), in three categories:

1 < IS < 3 poor fodder species; 4 < IS < 6 good fodder species; and 7 < IS < 9 very good fodder species. For each category, we calculated the rate of variations (loss or gain) and we also traced the biological spectra inside and outside the nine recording stations.

## Results and discussion

The stations planted were richer in species than the non-planted stations. This difference can be explained by the wet microclimate generated by the tufts of *Atriplex canescens*. First, this influences the pedo-climate positively by decreasing the action of the wind and thus reduces evapotranspiration so that the water reserves are conserved for the thérophytes especially to develop (Daget, 1980; Barbero *et al.*, 1990). Secondly, by trapping sand it allows the development of annual species by the phenomenon of self-mulching after storage water (Kadi-hanifi, 1998). At the level of the plantations, there is a predominance of thérophytes (Table 1).

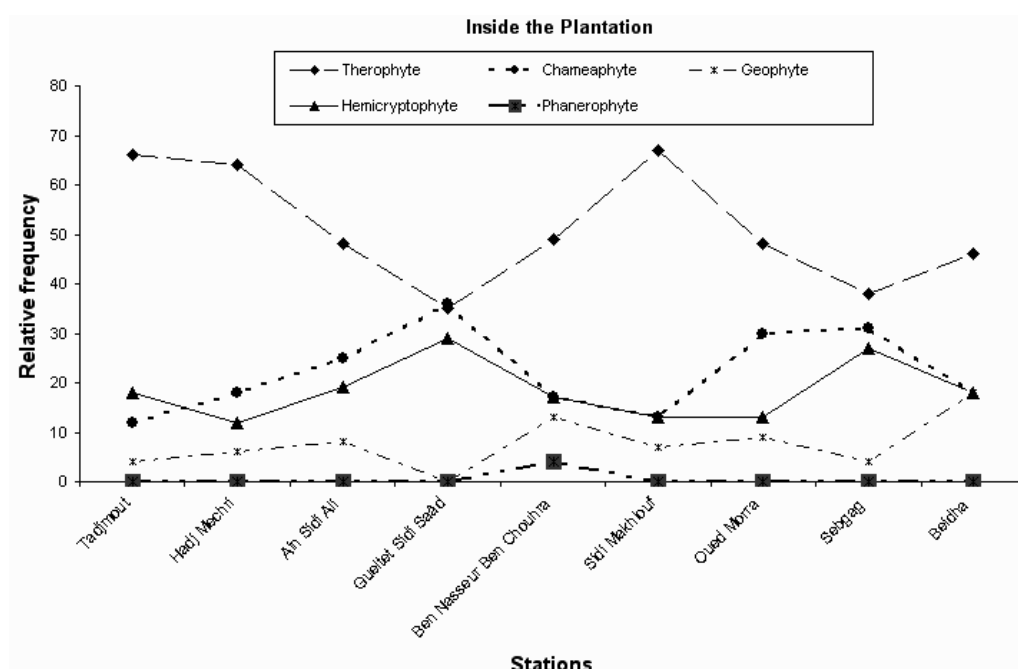
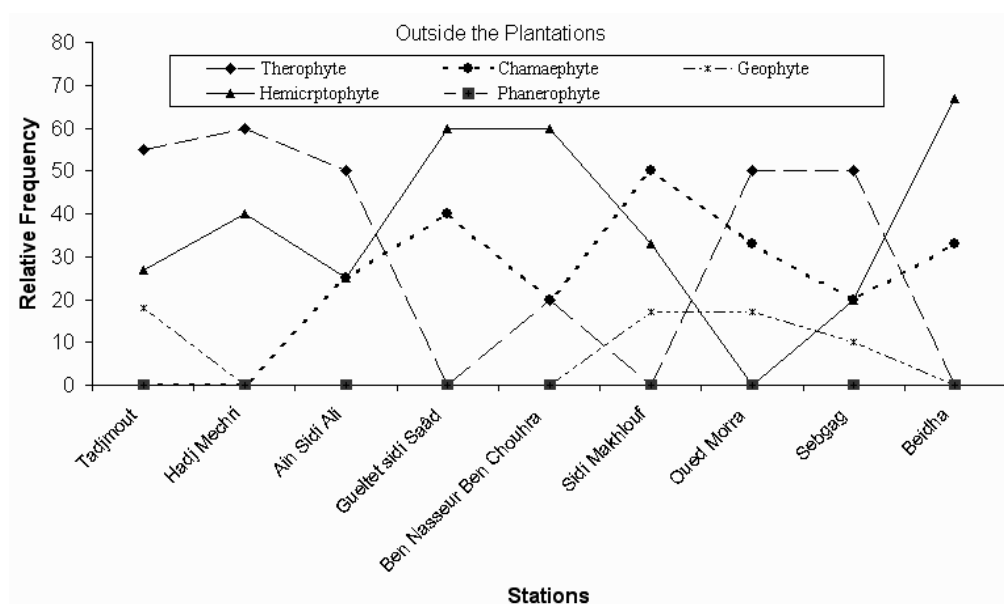
Table 1. Floristic richness, Sorensen index and rate of palatable species variation inside and outside the nine stations. A: Inside the plantation. B: Outside the plantation.

Stations	Number of species inside the plantation	Number of species outside the plantation	Number of common species	Index of Sorensen	Rate of pastorals species variation by pastoral class index		
					$\frac{2c}{a+b} \times 100$		
	a	b	C		1<Is<3	4<Is<6	7<Is<9
Tadjmout	51	11	10	+ 32.3	A	14	7
					B	4	2
Hadj Mechri	17	5	4	+ 36.4	A	4	2
					B	2	1
Ain Sidi Ali	36	4	2	+ 10	A	10	5
					B	1	0
Sidi Saâd	14	5	5	+ 52.6	A	3	1
					B	2	0
Ben Chouhra	23	5	4	+ 28.6	A	4	1
					B	2	0
Sidi Makhlouf	15	6	1	+ 9.5	A	3	3
					B	1	2
Oued Morra	23	6	2	+ 13.8	A	8	1
					B	3	1
Sebgag	49	10	6	+ 20.3	A	10	6
					B	1	0
Beidha	11	3	2	+ 28.6	A	1	2
					B	1	0

This therophytization is the consequence of self-mulching, which allows water storage and thus supports the development of annuals. At some stations outside the plantation there was a predominance of hemicryptophytes, which is explained by a significant rainfall during the winter and spring 2006. There was a negative correlation between the hemicryptophytes and the therophytes, which was also mentioned by Kadi-Hanifi (1998) and Amghar (2002).

## Conclusion

The systematic analysis shows an important diversity in families and an important generic and specific richness, the principal families represented are: *Asteraceae*, *Poaceae*, and *Fabaceae*. The analysis of the biological spectra shows the predominance of the thérophytes, species producing many seeds that colonize quickly open spaces.



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# Effects of agricultural practices on spatial dispersal patterns of permanent grassland species

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## Abstract

The effects of agricultural practices on floristic composition of grasslands are well known but few studies have investigated these effects on plant spatial patterns involved in the maintenance of plant diversity. In order to identify major processes controlling dispersal patterns of four grassland species, which represented different dispersal methods (anemochorous, barochorous and zoochorous), spatial analysis (4TLQV and Wavelet) was used and adapted to grid sampling. The quadrat variances were plotted against block size, and the peaks of graphs interpreted as the scale of the mapped phenomena. The results highlighted the general impacts of mowing in relation to the dispersal characteristics of different plant species. An effect of mowing on spatial patterns from functional traits connected to dispersal was shown by spatial analysis, e.g. dispersal distances of barochorous species such as *Ranunculus bulbosus* were enhanced by mowing and grazing and reached a maximum of 1 m, whereas a zoochorous grass (*Festuca arundinacea*), which has the theoretical capacity to produce expansive lateral spread up to 1 m, was limited in its expansion by cutting and grazing to 0.30 m. The study also showed that disturbance can play an important role in dispersal and the spatial structure of grassland species.

Keywords: spatial pattern, Four-Term Local Quadrat Variance, plant trait, dissemination, agricultural practices

## Introduction

The effects of agricultural practices on floristic composition of grasslands are well recognized and known to be related to plant traits. Plant traits are any morphological, physiological or phenological feature measurable at the individual level, from the cell to the whole-plant level, without reference to the environment or any other level of organization (Violle *et al.*, 2007). The identification of plant traits that explain responses of species to the intensity of grazing or mowing is one of the main tools in understanding the management of grazed systems (Weiher *et al.* 1999) but few studies have investigated the management-practice effects on plant spatial patterns involved in the maintenance of plant diversity. Spatial distributions in grasslands are mainly dependent on the vegetation propagation mode and seed dispersal. Species with structures that favour wind and animal dispersal have better regional and local-scale invasion success. At smaller spatial scales, further research is required on the determining factors of dispersal and colonization patterns. Indeed, the natural dispersal capacities of plant can be modified by the effect of agricultural practices. Here we explore the effects of agricultural practices on the plant spatial patterns of plant species representing different agents of dispersion, such as wind (anemochorous), gravity (barochorous) or animals (zoochorous).

## Materials and methods

We studied spatial patterns of one common grass (*Festuca arundinacea*) and three forbs (*Ranunculus bulbosus*, *Taraxacum officinale*, *Trifolium pratense*) chosen for their dispersal characteristics and lateral vegetative expansion capacities *sensu* Grime *et al.* (1988) (Table 1). *Festuca arundinacea* is mainly dispersed by animals and its theoretical lateral spread is up to 1 m; *T. officinale* is dispersed by wind and its lateral spread does not exceed 0.10 m, while *T. pratense* and *R. bulbosus* are grassland species with a barochorous dispersal mode with a lateral spread limited, respectively, to 0.25 m and 0.10 m. We established experimental grids on six permanent grasslands with a clay-loamy soil at the experimental installation INRA-SAD at Mirecourt, France (6°81' E, 53°6') in May 2007. Three grassland areas were grazed by dairy cows at a stocking rate of 1.5 UGB ha<sup>-1</sup> y<sup>-1</sup>, and three were mown twice a year. One experimental grid (4 m x 4 m) was randomly selected in each permanent grassland, and coverage of species was recorded in each of 1,600 (10 cm x 10 cm) quadrats.

In order to identify major processes controlling dispersal patterns of four grassland species, representing several modes of dispersal (anemochorous, barochorous and zoochorous, *sensu* Grime *et al.*, 1988), we used two spatial analyses: 4TLQV and Tall French Hat Wavelet, adapted to grid sampling (Dale, 1999). With these methods, when the quadrat variances are plotted against block size, the peaks of these graphs are interpreted as the scale of the mapped phenomena i.e. the size of the patch and the zone of inflection of the curve was the size of the gaps (Guo and Kelly, 2004; Perry I., 2006). All analyses were performed with the Passage package (Rosenberg, 2001).

Table 1. Dispersal capacities and spatial pattern analysis (for *Festuca arundinacea*, *Trifolium pratense*, *Ranunculus bulbosus*, *Taraxacum officinale*) under grazed and mown management.

Species	Dispersal agent	Theoretical spatial patterns expected	Lateral spread (m) <i>sensu</i> Grime <i>et al.</i> 1988	4 TLQV spatial patterns (m)		Tall French Hat Wavelet spatial patterns (m)	
				grazed	mown	grazed	mown
<i>F. arundinacea</i>	zoochorous	random	0.25-1.00	0.30	0.30	-	-
<i>T. pratense</i>	barochorous	aggregated	0.10-0.25	0.90	0.50	-	0.70
<i>R. bulbosus</i>	barochorous	aggregated	< 0.10	1.00	0.90	-	-
<i>T. officinale</i>	anemochorous	random	< 0.10	0.80	1.00	-	-

## Results and discussion

Impacts of mowing and grazing according to dispersal methods were highlighted by our results (Table 1). An effect of mowing and grazing on spatial patterns from functional traits connected to dispersal was shown by spatial analysis. However, the Tall French Hat Wavelet method does not give satisfactory results and does not allow the detection of the spatial patterns.

Dispersal distances of the barochorous species *R. bulbosus* and the unspecialized species *T. pratense* were enhanced by mowing and grazing. The size of the patch, highlighted by the spatial pattern analysis of *R. bulbosus*, increased and reached a maximum of 1 m under grazing management (Table 1). In the case of *T. pratense*, the patch size was greater under grazing conditions than under mowing management (0.90 m and 0.50 m, respectively; Figure 1). Grazing can locally reduce the above-ground competition with other species in the community by increasing the availability of light. *Trifolium pratense* responded by developing rhizomes under grazing conditions probably faster than in mown grasslands. In contrast, the zoochorous grass *F. arundinacea*, which has the theoretical capacities to produce

expansive lateral spread up to 1 m, was limited in its expansion by cutting and grazing to a patch size of around 0.30 m. The effect of grazing or cutting modified the random theoretical patterns expected by decreasing lateral expansion of *F. arundinacea* (Table 1). *Taraxacum officinale* had a theoretical random spatial pattern due to its mainly anemochorus dispersal method. Under management, the spatial pattern observed for *T. officinale* became aggregated and the patch size was greater than its lateral capacities (Table 1), so wind was not effective for the dispersal of seeds. The competitive pressure from surrounding tall grasses such as *Lolium perenne* or *Alopecurus pratensis* (data not shown) probably limits the dissemination capacities of *T. officinale*.

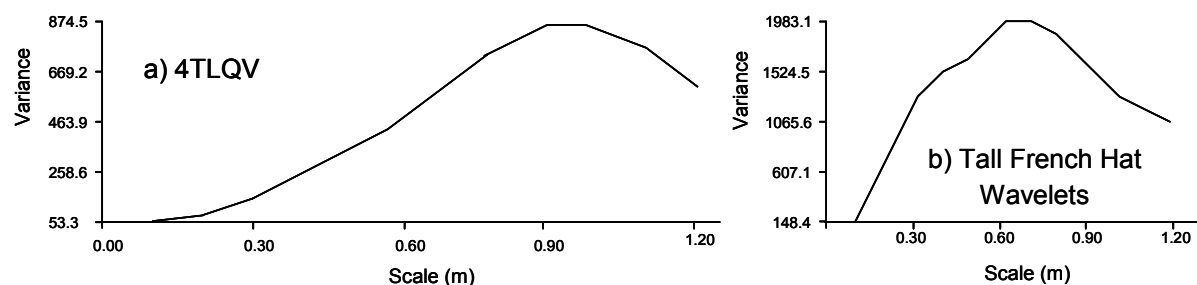


Figure 1. Spatial patterns for *Trifolium pratense* in grazed grasslands (a: 4TLQV) and mown grasslands (b: Tall French Hat Wavelets). The scale of window-width is plotted in units of quadrats (0.1 m).

## Conclusions

The analysis of the two spatial patterns performed in this study did not give similar results and their use must be complementary, but the Wavelet method does not allow the detection of spatial patterns. The size of the grid seems insufficient because the zone of inflection of the curve is not observed and this does not make it possible to determine the size of the gaps. However, this study showed that agricultural practices could play an important role in dispersal regime and in the spatial structure of grassland species. Grazing and mowing decrease or increase the size of the theoretical spatial patterns according to the dispersal characteristics of the species being considered.

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# Effect of different grazing systems on communities of epigeic beetles in a submontane area

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## Abstract

The effects on beetle communities of different grazing systems and management practices was studied using pitfall trapping on two pasture sites in the submontane area of Central Europe (South Bohemia). At site 1 the management was initial mowing and intensive grazing at 4 cows ha<sup>-1</sup>; site 2 had no mowing and less intensive grazing at 2 cows ha<sup>-1</sup>. Ordination by DCA analysis for the comparison of both sites, and direct RCA analysis for the evaluation of management effects on the species, were used for statistical evaluation of the data on beetle numbers and species. The effect of agricultural management was studied by determining the frequency of the species of different ecological groups. The number of species discovered by pitfall trapping was practically the same in both pastures. Intensive grazing resulted in the extinction of some stenotopic and hygrophilous beetle species, whereas the proportion of polyphagous and eurytopic species was higher. The effect of less intensive grazing with fewer animals resulted in the mosaic of vegetation which is favourable for some stenotopic epigeic beetles (e.g. some large species of the genus *Carabus*). Some rare species living in dung (e.g. staphylinid beetle *Philonthus marginatus*) were present in both pasture types. Invasive species were found only in the pasture with more intensive management.

Keywords: grazing intensity, epigeic beetles, communities, Central Europe

## Introduction

Epigeic beetles, especially carabids and staphylinids, are extremely diverse and distributed in all types of terrestrial ecosystems (Bohac, 1999; Holland, 2002). They play an important role in the cultural landscape as predators of pests. More recently, they have been used as bio-indicators responding to chemical pollutants (e.g. pesticides and heavy metals) or to management regimes (e.g. of crops, moorland and grassland). It is known that intensive grazing decreases the occurrence of the stenotopic species (species with narrow ecological niche living only in special biotopes) of epigeic invertebrates in grasslands, with an increasing number of species having characteristics pertaining to field or eurytopic species (species with a wide niche living in a wide range of biotopes) (Mladek *et al.*, 2006). On the other hand we have insufficient information about the effect of different types of pasture management on separate groups and species of invertebrates. The aim of this paper was to find out whether different intensities of grazing and varied management practices have an essential influence on beetle communities in submontane areas in the Czech Republic.

## Materials and methods

The effect of different grazing systems and different management of two pastures on the beetle communities was studied using pitfall trapping in the submontane area of central Europe (South Bohemia). Detailed information on the pastures is given in Table 1.

A row of 20 pitfall traps (diameter 7 cm) was installed in each plot. Pitfall traps were filled with ethylene glycol. The material from the traps was collected every month from April to October 2006.

The program CANOCO version 4.51 was used for the statistical evaluation of the pitfall trap data; graphical outputs were elaborated by the CANODRAW and CANOPOST programs (ter Braak and Šmilauer, 1998). We used DCA analysis for the comparison of sites and direct RDA analysis for the evaluation of management effect on the species. For more transparency of the graphical outputs, 21 species were visualized. The intensity of management was tested in the model. The degree of human impact was studied by the frequency of the species of different ecological groups (Boháč, 1999). The method of ecological analysis of beetle communities (Boháč, 1999) was used for evaluating the community structure.

Table 1. Characteristics of surveyed pastures.

	Farm 1	Farm 2
Altitude	700-790 m a.s.l.	740-789 m a.s.l.
Duration of grazing season in 2004	171 days	174 days
Duration of grazing season in 2005	146 days	151 days
Stay at pasture sites	day and night	in shed during the night
Number of cows per ha	4 cows per ha	2 cows per ha
Continuity of pasture at the locality	15 years	20 years
Initial mowing	no	yes
Number of plant species	15	23

## Results and discussion

The number of species discovered by pitfall trapping in pastures with different management was practically the same: 76 from the pasture with less-intensive grazing, and 74 from the pasture with more-intensive grazing). Intensive grazing in the pasture resulted in the extinction of some stenotopic and hygrophilous species (e.g. some ground beetles and staphylinid beetles). The proportion of polyphagous species was greater in the pasture with more intensive grazing (43%) in comparison with the other plot (26%). The invasive species of beetles were found in the pasture with more intensive grazing (e.g. staphylinid beetle *Philonthus spinipes*). The effect of less-intensive grazing with a lower number of cows resulted in the mosaic of vegetation which is favourable for some stenotopic epigeic beetles (e.g. some large species of the ground beetles, e.g. *Carabus granulatus*). Some rare species living in dung (e.g. the staphylinid beetle *Philonthus marginatus*) were present in both pasture sites with different grazing intensity and management.

Ordination of beetle samples from the studied pastures with different grazing regimes and management carried out by DCA showed considerable similarity of both management variants (the first axis explains 47.1% of variability, the second 9.2% of variability). On the other hand, there are some distinct differences in the species composition in both variants. The pasture with more intensive grazing is characterized by species tolerant of drier soils (e.g. the carabids *Harpalus rubripes* and *Amara plebeja*), coprophagous species (e.g. *Aphodius rufipes*) and eurytopic phytophagous species tolerant of short vegetation (e.g. *Psylliodes affinis*). The pasture with combined management, with lower intensity of grazing, hosts species with higher hygropreferendum (e.g. *Dryops* spp.) and with preference of sandy and less compact soils (e.g. the carabid *Clivina fossor*).

Ordination of beetle species by PCA (Fig. 1) indicates the dominance of coprophagous species (e.g. the species of genera *Aphodius* and *Cercyon*) and eurytopic species (e.g. the species of genera *Bembidion*, *Amara*, *Philonthus* and *Quedius*) on pastures with more intensive grazing. The hygrophilous species and species with preference of shaded biotopes are characteristic of the grassland with less intensive grazing (e.g. the carabid beetle *Chlaenius nigricornis*, and the staphylinids *Quedius fuliginosus*, *Ocypus tenebricosus*, etc.). There is no significant influence of the different management and grazing regime (more intensive in the first pasture – 4 cows ha<sup>-1</sup> and less intensive in the second pasture – 2 cows



ha<sup>-1</sup>; initial mowing of the second pasture, absence of mowing in the first pasture) on the beetle communities ( $F = 3.038$ ,  $P = 0.068$ ).

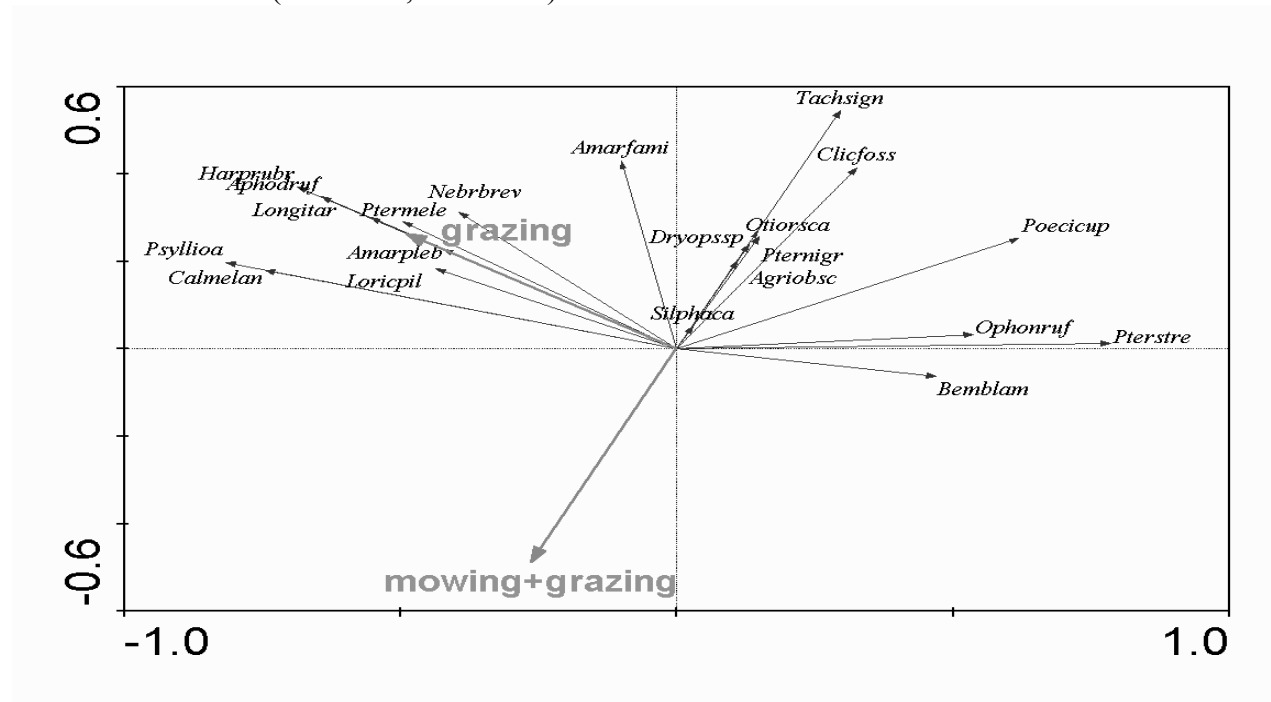


Figure 1. Ordination of beetle species by PCA (activity of species is increasing in the direction of arrow.) The abbreviations of beetle species see text.

## Conclusions

Based on the results of analysis of beetle communities in two submontane pasture sites with different intensities of cattle grazing, it can be concluded that grazing at 4 cows ha<sup>-1</sup>, compared with an intensity of 2 cows ha<sup>-1</sup>, has a relatively small effect on the total number of beetle species present. On the other hand, the eurytopic, and even invasive species, were more active in the site with intensive grazing. This result indicates the possibility of the penetration of eurytopic and invasive beetle species to submontane landscapes, namely grasslands with intensive cattle grazing. The results need to be tested for additional combinations with different environmental conditions.

## Acknowledgements

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# Relationship between vascular plant species richness and soil chemical properties of alpine meadows and pastures

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## Abstract

In unfertilized mountainous grassland communities in the Austrian Alps, there is a hump-shaped or unimodal relationship between vascular plant species richness and soil pH (pH-CaCl<sub>2</sub>). Only the combination of nutrient-poor grassland soil, minimal stress (well-drained grassland soil in the silicate buffer range) and moderate disturbance (periodical or episodic mowing, extensive grazing) in the plant habitat, and a high regional species pool is associated with maximum species richness. There is, however, no relationship between vascular plant species richness and the commonly used indicators for forage quality.

Keywords: alpine meadows and pastures, vascular plant species richness, soil chemical properties, calcifuges, calcicoles, forage quality

## Introduction

Grassland communities differ not only in species composition, but also in vascular plant species richness, yield, and forage quality. The aims of this study were (1) to investigate the significance of soil chemical properties in influencing vascular plant species richness, (2) to examine the influence of soil chemical properties on the distribution of the calcifuge and calcicole species, and (3) to investigate the relationship between vascular plant species richness and forage quality.

## Materials and methods

This investigation was carried out in unfertilized mountainous grassland communities in the Austrian Alps on 42 different sites, distributed over Carinthia. To determine vascular plant species richness (alpha-diversity), the total number of vascular plant species within a homogenous investigation area of 50 m<sup>2</sup> was recorded. Only unfertilized and extensively used montane, subalpine, and alpine meadows and pastures were investigated and only vascular plants were taken into consideration. The altitude ranged from 1,340 to 2,220 m a.s.l. Soil moisture regime was generally well balanced or periodically moist in topsoil. The vegetation types considered in this study belong to the *Nardo-Agrostion tenuis*, *Nardion strictae*, *Festucion varia*, *Caricion curvulae*, *Caricion ferrugineae*, and *Seslerion caeruleae* (Grabherr and Mucina, 1993; Mucina *et al.*, 1993). These are the most widespread semi-natural grassland communities in the Austrian Alps and they represent a vegetation and soil gradient from plant communities on very acid soils to grasslands on neutral and alkaline soils.

Soil samples were collected from the 0-10 cm soil layer (A horizon) in the vegetation period. From field-moist soil samples, a saturation extract was produced according to ÖNORM L 1092-93 (Austrian Standards). Ions were analysed by ICP and ion chromatography. Dissolved organic carbon (DOC) was determined by oxidation with K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>-H<sub>2</sub>SO<sub>4</sub>. Mineral element content in the harvestable above-ground plant biomass and forage quality (crude fibre, crude protein, crude fat, crude ash, digestibility of organic matter, net energy) were analysed by using standard methods. SPSS (Version 14.0) was used for all statistical data analyses.

## Results and discussion

Across all sites investigated, species richness varied between 16 and 96 vascular plant species per 50 m<sup>2</sup>, and soil pH(CaCl<sub>2</sub>) ranged from 3.4 to 7.6. Especially the grassland communities on soils formed from calcareous mica schists have the greatest plant species richness. There was a hump-shaped or unimodal relationship between vascular plant species richness and soil pH (Figure 1).

Tyler (2003) also observed in non-fertilized grasslands in south-eastern Sweden that the highest number of plant species per 25 m<sup>2</sup> was in the intermediate pH range and lower values at pH-KCl 4 and 7.5. According to Ulrich (1981) the soils can be grouped into different buffer ranges. Statistical analyses (Kruskal-Wallis test, Mann-Whitney test) revealed, that highly significant differences ( $P < 0.005$ ) between the grouped soils in Table 1, in terms of mineral element concentrations in the saturation extract, were restricted to P, Si, K, Ca, Zn, and Cu.

Table 1. Mean ion concentrations in the saturation extract; n = number of soil analyses.

pH-CaCl <sub>2</sub> buffer range	n	P	Si	K	Ca	<sup>mg L<sup>-1</sup></sup> Zn	Cu	Al	Fe	Mn
< 4.2	22	0.72	2.35	2.64	5.16	0.07	0.01	0.51	0.52	0.07
4.2-5.0	9	0.20	1.31	2.78	7.23	0.03	0.01	0.36	0.16	0.24
5.0-6.2	8	0.26	1.45	1.41	12.49	0.05	0.03	0.43	0.54	0.09
> 6.2	5	0.38	1.71	1.42	38.22	0.02	0.02	0.65	0.53	0.07

Table 2. Mean molar ratios in the saturation extract.

pH-CaCl <sub>2</sub> buffer range	Ca:K	Mg:K	Ca:Mg	Ca:Al	Mg:Al	C:Al	C:(Al+Fe)
< 4.2	2.4	0.8	3.0	8.3	2.8	177	132
4.2-5.0	4.0	1.2	3.2	14.6	5.6	368	306
5.0-6.2	13.0	4.8	2.6	100.6	55.9	719	356
> 6.2	45.0	12.7	10.5	93.5	9.1	415	249

Very acidic alpine topsoils in the aluminium and/or iron buffer range (pH-CaCl<sub>2</sub> < 4.2) are characterized by a relatively high concentration of P, Si, K, and Zn in the soil solution, primarily due to an intensive chemical weathering and mineral dissolution (Table 1), by a relative excess of K- and Al-ions, and by a complementary lack of Ca- and Mg-ions in the soil solution (Table 2). Furthermore, these soils have a comparatively high mean NH<sub>4</sub>-N:NO<sub>3</sub>-N ratio of 69 in the LiCl-exchangeable fraction, compared with soils in the silicate buffer range (pH-CaCl<sub>2</sub> 6.2-5.0) or carbonate buffer range (pH-CaCl<sub>2</sub> > 6.2) with ratios of 32 and 11, respectively. Only calcifuge species are adapted to the prevailing acid and nutrient stress mainly caused by the unfavourable ratios between nutrients as well as potentially toxic elements (e.g. Al) in the soil solution. Especially the uptake of Ca and Mg by plants seems to be inhibited, whereas the uptake of Na, Fe, Mn, Zn, Cd, and Cr might be promoted (data not shown). Therefore, vascular plant species richness is very low, and calcifuges are dominating the species composition of the vegetation. Al-toxicity *per se* appears not to influence the vascular plant species richness, because humus-rich alpine topsoils have a relatively low Al-concentration in the soil solution (Table 1). Therefore, the molar ratios of Ca:Al, Mg:Al, and C:Al are relatively wide (Table 2). Furthermore, the excess of DOC promotes the formation of complexes of Al with DOC and, hence, Al-detoxification. Only in extremely acidic soil solutions does this form of Al-detoxification become ineffective, and Al occurs mainly as toxic, monomeric Al<sup>3+</sup> in the soil solution. On the other hand, calcareous soils in the carbonate buffer range are characterized by an absolute and relative excess of Ca-ions in the soil solution (Table 1 and 2). Only calcicole species are adapted to this disharmonic nutrient supply and the resulting discrimination of individual nutrients. Especially the uptake of P, S, Mn, Zn, and Cu by plants seems to be inhibited, whereas the

uptake of Ca might be promoted (data not shown). Therefore, vascular plant species richness is moderately high, and calcicoles are prevailing. Soils in the silicate buffer range and soils in the upper part of the cation exchange buffer range (cation exchange buffer range: pH-CaCl<sub>2</sub> 5.0-4.2) are generally characterized by a balanced composition of the soil solution (Table 1 and 2). Therefore, both calcifuges and calcicoles can be present at these sites, allowing a high vascular plant species richness. There was, however, no relationship between vascular plant species richness and commonly used indicators for forage quality such as crude protein, crude fat, crude ash, digestibility of organic matter or net energy (unpublished data).

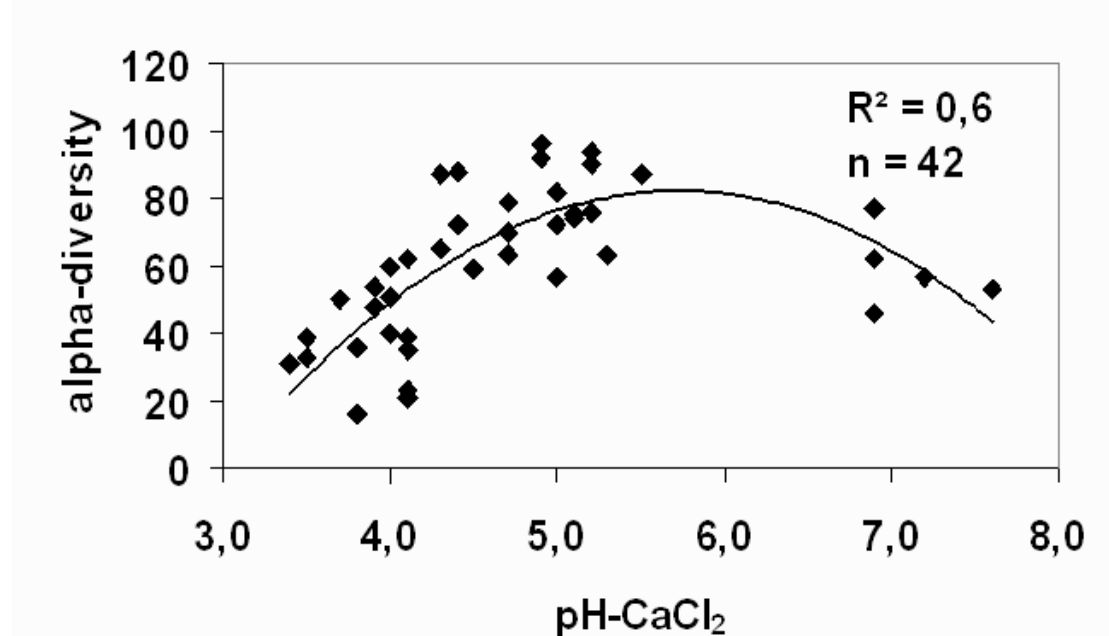


Figure 1. Relationship between vascular plant species richness per 50 m<sup>2</sup> and soil pH.

## Conclusions

In unfertilized mountainous grassland communities in the Austrian Alps, the pH-dependent soil chemical properties in topsoil are of utmost importance to vascular plant species richness. Nutrient-poor plant habitats are potentially rich in vascular plant species if the grassland soils are in the silicate buffer range or in the upper part of the cation exchange buffer range. They are particularly poor in vascular plant species if the grassland soils are in the aluminium and/or iron buffer range.

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# The effect of sowing date, cover and catch crops on the productivity of resown pasture swards

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## Abstract

Pasture sward improvement investigations were conducted during the period 1980-2003 at the Vėžaičiai Branch of the Lithuanian Institute of Agriculture. The focus was on the effects of pasture turf cultivation and resowing time, different cover and catch crops, and herbicide use in combination with pasture resowing. Cover crops, catch crops and pasture resowing time all affected dry matter (DM) yield. The most suitable time for turf cultivation was August-September and spring was most suitable for grass sowing. The best cover crops were barley for grain and oat-vetch mixture for green forage/silage. Potato and cereals were the best catch crops for pasture resowing. DM yield of resown pasture increased by 0.04-2.18 t ha<sup>-1</sup> compared with old untreated pasture. Resowing led to increased proportions of legumes in the sward by 1.1-9.2%, while the proportions of forbs decreased by 9.1-14.9%.

Keywords: pasture resowing, cover and catch crops, productivity, botanical composition

## Introduction

For sufficient feed production farmers need to establish high-quality and productive pasture swards. Therefore, pasture sward improvement is particularly important. Previous research (Slamka *et al.*, 1999; De Vliegher *et al.*, 2002; Butkuvienė, 2006) has shown that pastures poor in botanical composition and productivity need to be resown. Pastures are usually resown using a method in which the old pasture turf is destroyed in autumn, and a seed mixture is then sown in the following spring, without catch crops (Daugėlienė, 2002). Pasture swards are also resown using catch crops. Cover crops are necessary both for resowing pastures without or with catch crops. Lithuanian researchers (Zimkus, 1988; Butkuvienė, 2006) have determined that the best cover crops for pasture resowing are barley for grain and oat-vetch mixture for green forage or silage. The aim of this study was to evaluate the effects of the main improvement methods on sward productivity and botanical composition.

## Materials and methods

Pasture sward improvement investigations were conducted on grass-dominant pastures of 6-18 years old. White clover (*Trifolium repens* L.) accounted for 5-20% and forbs for 15-30%. The research was carried on Haplic-Albic Luvisols with a topsoil pH<sub>KCl</sub> 5.2-6.2; soil P was 60-220 mg P<sub>2</sub>O<sub>5</sub> kg<sup>-1</sup>; soil K was 104-115 mg K<sub>2</sub>O kg<sup>-1</sup> and humus was 2.5-3.8%. Four groups of research treatments were carried out: i) pasture turf cultivation and resowing time (N<sub>120</sub>P<sub>60</sub>K<sub>60</sub>); ii) cover crops selection and harvesting time (N<sub>60</sub>P<sub>60</sub>K<sub>60</sub>); iii) catch crop selection (N<sub>90</sub>P<sub>60</sub>K<sub>60</sub>); and iv) herbicide use in combination with pasture resowing (P<sub>60</sub>K<sub>60</sub>). Details are summarized in Tables 1-4. The same seeds mixture was used for resowing in all trials. It contained white clover cv. 'Atoliai' 25%, timothy (*Phleum pratense* L.) cv. 'Gintaras II' 40%, smooth-stalked meadow grass (*Poa pratensis* L.) cv. 'Danga' 25% and meadow fescue (*Festuca pratensis* Huds.) cv. 'Dotnuvos I' 10%. Plot size was 3 m x 15 m. Four experiments were carried out, each in a randomized design with four replicates. Assessments were made of herbage production in terms of dry matter (DM) yield and metabolizable energy (ME) value.

## Results and discussion

*Effects of time of cultivation and time of sowing.* There was a degree of similarity in terms of DM yield and ME between the treatments that were cultivated in autumn (August-September) and then sown in spring: 7.12-7.28 t DM ha<sup>-1</sup> and 73.3-75.0 GJ ME ha<sup>-1</sup> respectively for August and September cultivations (Table 1). The treatments with cultivation in May-June, followed by sowing before 1 July, resulted in the same sward productivity. Cultivating the turf and resowing later in the summer resulted in decreased pasture productivity relative to the other sown treatments. The lowest DM yield and ME production (5.14 t DM ha<sup>-1</sup> and 52.9 GJ ME ha<sup>-1</sup>) was obtained when the old sward was cultivated at the end of July and resown within a month. Cultivation and resowing, except the treatment with the latest sowing date, resulted in considerably increased proportions of legumes in the swards, compared with the unsown old pastures. Resowing significantly decreased the proportion of forbs in the sward.

Table 1. The effect of time of cultivation and resowing on pasture productivity and botanical composition.

Turf cultivation time	Sowing time after turf cultivation	DM yield t ha <sup>-1</sup>	ME GJ ha <sup>-1</sup>	Legumes, %	Forbs, %
Old pasture		5.10	52.5	6.7	14.5
May	after 2 weeks	6.89	70.0	11.7	5.4
	after 4 weeks	7.05	72.6	11.8	5.2
June	after 2 weeks	7.06	72.7	13.2	6.0
	after 4 weeks	6.56	67.0	10.8	6.2
July	after 2 weeks	6.18	63.6	11.2	6.8
	after 4 weeks	5.14	52.9	5.5	7.5
August	in spring	7.12	73.3	12.2	5.2
September	in spring	7.28	75.0	12.9	4.5
October	in spring	6.84	70.4	11.6	5.4
LSD <sub>05</sub>		0.55	3.7	5.7	3.5

*Cover crop selection and harvesting time.* The greatest amount of additional pasture DM yield (0.54 t ha<sup>-1</sup>) was obtained when the cover crop, barley, was harvested at complete maturity (Table 2). Slightly less (0.26-0.31 t DM ha<sup>-1</sup>) additional pasture yield was obtained when the oat-vetch mixture was grown for green forage or silage. Nevertheless, oat for grain was not found to be a suitable cover crop, as no additional DM yield was obtained. Analysing the ME data showed an analogous trend to the DM yield results. The influence of cover crop on botanical sward composition was observed only in the first year of use. The effect of treatment on the proportions of legumes and forbs was small.

Table 2. Influence of cover crop on productivity and botanical composition of resown pasture.

Treatments	DM yield, t ha <sup>-1</sup>	ME, GJ ha <sup>-1</sup>	Legumes, %	Forbs, %
Without cover crop	6.23	63.8	21.4	5.1
Vetch-oat mixture for green forage	6.53	66.3	21.0	5.8
Vetch-oat mixture for silage	6.48	66.9	20.4	4.9
Barley harvested at complete maturity	6.77	68.9	20.9	4.3
Barley harvested with 2 weeks delay	6.32	64.3	19.5	4.5
Oat for green forage	6.38	65.9	20.8	4.6
Oat for grain	5.97	61.8	21.2	4.7
LSD <sub>05</sub>	0.10	1.3	2.9	1.1

*Catch crop selection.* Table 3 shows that, on average for the year of the catch crop introduction, the highest amount of ME (43.5 GJ ME ha<sup>-1</sup>) accumulated when pastures were resown after a catch crop for green forage and insignificantly less (42.3 GJ ME ha<sup>-1</sup>) after a

potato crop, which had been manured at 60 t ha<sup>-1</sup>. The pastures resown without a catch crop or after cereals resulted in almost the same ME accumulation, 34.8 and 34.1 GJ ME ha<sup>-1</sup> respectively. Pasture DM yield was greater when pasture was resown after a catch crop. The greatest amount of additional pasture DM yield (0.58 t DM ha<sup>-1</sup>) was obtained after potatoes. Pasture resowing, with or without catch crops, significantly increased legume proportion and decreased the proportion of forbs in the sward.

Table 3. Effects of different catch crops on resowing of pastures.

Treatments	Pasture DM yield, t ha <sup>-1</sup>	ME, GJ ha <sup>-1</sup>	Legumes, %	Forbs, %
Non-resown pasture	2.78	30.8	12.4	27.9
Without catch crops	2.90	34.8	20.1	18.4
Catch crop - potato	3.36	42.3*	21.6	13.0
Catch crop - cereals	3.11	34.1*	20.3	14.2
Catch crop - green forage crops	3.07	43.5*	20.5	15.3
LSD <sub>05</sub>	0.09	0.68	6.0	2.6

\* - pasture + catch crop ME

*Herbicide use in combination with pasture resowing.* Research was carried out on different pastures: i) sprayed with herbicide MCPA (at 3.7L ha<sup>-1</sup>) in autumn, at the beginning of the research, and ii) not sprayed with herbicide. Pasture resowing significantly increased DM yield by 1.45-1.68 t DM ha<sup>-1</sup> (Table 4). Significantly higher amounts of ME were obtained when improving pasture by resowing with legume-grass mixture in both sprayed and non-sprayed pastures. The average results showed that the sward was enriched with legumes by 6.4-7.9% after pasture resowing and the amount of forbs was significantly decreased by 9.4-11.3%.

Table 4. Effect of agricultural methods (with/without herbicide) for improving pasture swards.

Treatments	DM yield, t ha <sup>-1</sup>	ME, GJ ha <sup>-1</sup>	Legumes, %	Forbs, %
Old pasture	3.66	38.3	18.4	28.8
Non-sprayed, resown pasture	5.11	55.8	24.8	19.4
Sprayed, resown pasture	5.34	57.3	26.3	17.5
LSD <sub>05</sub>	0.11	9.48	5.75	5.45

## Conclusions

Proper selection of pasture resowing time, cover crop and, if necessary, catch crop resulted in additional DM yields of up to 2.18 t ha<sup>-1</sup>. All the resowing methods considered resulted in an improved botanical sward composition and the forage quality was also increased.

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# Efficiency of environmental improvements in open areas of the Central Apennines Mountains (Italy)

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## Abstract

The aim of this work was to enhance the quality and productivity of habitats utilized by wild animals, such as deer and wild boar, by carrying out environmental improvements in open areas located in the Regional Park of Laghi di Suviana e Brasimone in Bologna, Italy. In the studied area shrubs and bracken fern (*Pteridium aquilinum*) were removed and a suitable forage mixture was seeded. Vegetation was sampled through linear analysis from which measures of botanical composition, pastoral value and biodiversity of the meadow were calculated. During the vegetation survey, the defoliation rate of the eaten species was also collected in order to obtain more information about the palatability of species. The vegetation changed considerably after the treatment, with significant enhancement in biodiversity, as measured by Shannon-Wiener index which increased from about 1.8 to 2.3. Pastoral value also showed a value triple that at the beginning of the trial. Moreover, the method used to measure the defoliation rate enabled effective indications to be made concerning the real utilization of several plant species by wild animals.

Keywords: environmental improvement, biodiversity, deer, pasture, defoliation rate

## Introduction

Agricultural abandonment in Europe reflects a post-war trend of rural depopulation. In many areas this has led to a decline in traditional practices and to an abandonment of marginal agricultural lands. Many studies show that in the absence of management (mowing, grazing, etc.) some undesirable plant species can expand, reducing the suitability of pastures for domestic livestock and for wild animals such as red deer (*Cervus elephas*), roe deer (*Capreolus capreolus*) and wild boar (*Sus scrofa*). One of the most invasive weeds is bracken fern (*Pteridium aquilinum*) (Le Duc *et al.*, 2000) whose presence reduces the qualitative value of the pastures for livestock and other fauna (Pakeman and Marrs, 1992; Cervasio *et al.*, 2007). The ratio between open pastures and woodland strongly influences the use of habitat resources by ungulates, with possible consequences for damage to the nearby crops (Danilkin, 1996). This study examined the effectiveness of agronomical management systems against bracken fern infestation in open areas, once used as pastures, situated in the Regional Park of Laghi di Suviana e Brasimone (Central Apennines, Bologna, Italy).

## Materials and methods

The trial site 'Lamaccia' is a 2.5 ha open area located at about 1100 m asl in the Regional Park of Laghi di Suviana e Brasimone. In the area environmental improvements included cutting of all invasive vegetation (41% bracken fern), and ploughing and sowing of a forage species mixture (30% *Bromus inermis*, 30% *Dactylis glomerata*, 25% *Festuca ovina*, 10% *Trifolium pratense*, 5% *Lotus corniculatus*). In the following years the sward was cut to maintain the new vegetation. Data on bracken cover and on botanical composition (8 samples per year) were recorded in summer months from 2004 to 2007 using linear analysis according



to the Daget and Poissonet method (1969). Botanical analysis permitted the evaluation of the specific contribution (SC), i.e. the percentage of each species in the total of the vegetation, and the pastoral value (PV) calculated using the formula  $PV = \sum (SC_i \times SI_i) / 5$ , where  $SI$  is a specific index, variable from 0 to 5, which summarizes the forage value of each species in the pasture (Cavallero *et al.*, 2002). Floristic richness was evaluated using the Shannon-Wiener index  $H' = - \sum p_i \ln p_i$ , where  $p_i$  is the percentage of the specific frequency in decimal fraction (Magurran, 2004). To analyse the effective use of the new pasture by wild ungulates and the direct impact on the different species, the defoliation rate (DR) was estimated according to Orth *et al.* (1998). The pastoral value, the Shannon-Wiener index and the species-richness during different years were analysed by ANOVA.

## Results and discussion

A large decrease in bracken specific contribution (SCB) can be noticed in the years after the agronomical management that was carried out in 2004 (Figure 1A). Moreover, establishment of the species of the sown sward mixture (SCM) and in the contribution of spontaneous species (SCS) was observed. The increased number of botanical families of pastoral interest after the agronomical treatments indicated an improvement of the pasture quality, shown by a significant increase of the pastoral value trend (Figure 1B).

Trends of biodiversity ( $H'$ ) and of species number ( $R$ ) express a floristic diversity that might be of interest for the wild fauna (Figure 2).

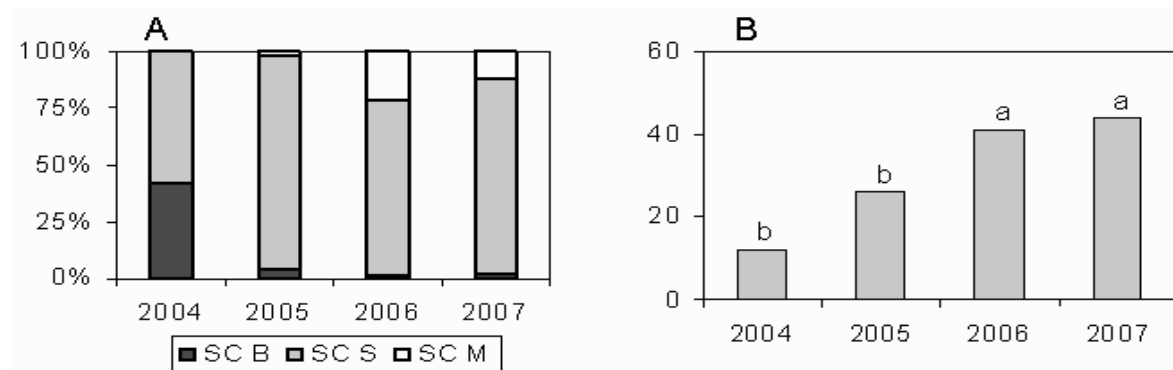


Figure 1A. Percentage of specific contribution of sward mixture (SCM), spontaneous species (SCS) and bracken fern (SCB). Figure 1B: Evolution of pastoral value (PV), along years of trial. Columns with different letters are significantly different at  $P < 0.001$ .

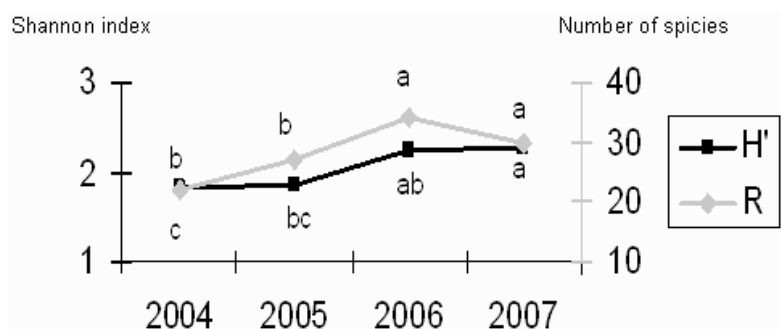


Figure 2. Shannon-Wiener index ( $H'$ ) and floristic richness ( $R$ ) during 2004-2007 period. Data with different letters along the same line are significantly different at  $P < 0.001$ .

Table 1 shows the utilization of some species during the summer months, expressed as the ratio between the defoliation rate (DR) and the specific contribution of the same species, (DR/SC). When this ratio is greater than 1 the species is considered as preferred by grazing ungulates. Table 1 also shows the specific index (SI) of each plant. It is interesting to notice that in the studied pasture, used only by wild animals, for many species there was not a strong relation between SI, usually used to calculate PV for livestock, and the parameter DR/SC. In fact species with SI low or equal to 0 were selected and utilized by the wild fauna, as shown by the correspondent values of DR/SC when higher than 1.

Table 1. Utilization rate (DR/SC) of some species grazed by the wild fauna and respective SI.

Species	DR/SC ratio					Species	DR/SC ratio				
	Jun	Jul	Aug	Sep	SI		Jun	Jul	Aug	Sep	SI
<i>Achillea millefolium</i>	1.89	1.05	2.94	1.03	2	<i>Leucanthemum vulgare</i>	3.16	-	-	-	0
<i>Carex otrubae</i>	-	-	-	3.05	0	<i>Plantago major</i>	-	-	-	2.28	1
<i>Cytisus scoparius</i>	-	0.66	9.71	-	0	<i>Prunella vulgaris</i>	-	3.54	-	-	0
<i>Festuca ovina</i>	-	-	-	2.48	1	<i>Ranunculus acris</i>	-	1.78	-	1.70	0
<i>Galium cruciata</i>	0.98	1.49	1.65	0.97	0	<i>Rumex acetosella</i>	1.12	1.32	-	0.85	0
<i>Galium palustre</i>	1.84	0.64	1.89	1.85	0	<i>Stellaria graminea</i>	0.46	1.11	0.46	0.35	0
<i>Holcus mollis</i>	1.46	1.32	1.93	1.16	2	<i>Stellaria media</i>	-	1.87	-	-	0
<i>Hypericum perforatum</i>	-	-	1.67	-	0	<i>Veronica chamaedrys</i>	1.21	0.63	2.10	1.37	0
<i>Knautia arvensis</i>	-	-	-	1.11	0	<i>Veronica officinalis</i>	-	-	-	2.73	0
<i>Leontodon hispidus</i>	-	-	-	4.93	1	<i>Viola tricolor</i>	1.03	1.80	1.75	3.01	0

## Conclusions

The results confirmed the effectiveness of the environmental improvement carried out, and also in the increasing of spontaneous botanic species. Management of open areas through a continuous programme of pasture maintenance can represent an important action for a correct exploitation of the ecosystem, preserving the floristic richness and consequently habitat biodiversity, and through making the pastures a stable food resource for fauna. This can mean positive outcomes on the management for fauna and in reduced damage to crops caused by wild ungulates in the surrounding agricultural areas.

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# Promoting biodiversity on intensively managed grassland in Scotland

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## Abstract

There is a need to combine agricultural production with the enhancement of biodiversity in intensively managed grasslands. The sympathetic management of field margins is one way Scottish farmers can integrate economic and environmental objectives. This includes sowing bird cover crop in the field margins, fencing off outer field margins to create conservation headlands and installing riparian buffers (i.e. fenced off areas adjacent to watercourses) to mitigate diffuse pollution by preventing livestock entering the watercourse. While such riparian buffers are becoming more common in Scotland their role in promoting biodiversity is largely undervalued. Current studies are examining a range of water margins to determine if sympathetically managed buffers enhance biodiversity in terms of plant, invertebrate and bird activity. Initial findings indicate that such buffers enhance several groups of invertebrates that are important prey items of farmland birds e.g. sawfly larvae, *Cicadellidae* bugs and harvestmen. Appropriately managed riparian margins in intensively managed grassland therefore have the potential to increase biodiversity at the field level, by increasing the abundance of key invertebrates, and at the landscape level, by increasing habitat heterogeneity.

Keywords: Riparian buffers, farmland biodiversity, intensive grassland, *Carabidae*

## Introduction

The intensification of farming practices over the past 50 years has led to homogenization of the agricultural landscape and increased disturbances at the field level which have in turn resulted in a steady decline in farmland biodiversity (Benton *et al.*, 2002). Agrienvironment schemes in Europe aim to halt and ultimately reverse this decline. Managing the outer margins of intensively managed fields for conservation has been shown to benefit biodiversity within arable landscapes (Siriwardena *et al.*, 1998) and initial findings suggest that similar benefits may also be possible in intensive grassland (Cole *et al.*, 2007; Woodcock *et al.*, 2007). The placement of such margins along riparian zones would not only target areas naturally rich in biodiversity, but also help to mitigate diffuse pollution. The installation of riparian buffers in intensive grassland is becoming more widespread in Scotland as a means of protecting the watercourse from pollutants associated with intensive livestock production. Through an examination of a range of riparian zones in intensively managed grassland this study aims to determine if such riparian buffers have an additional role to play in promoting farmland biodiversity.

## Materials and methods

Twenty-two locations were chosen from seven dairy farms in Ayrshire, Scotland (UK National Grid Reference: NS53) and investigated over a three-year period (2004-2006). Each location was classed as either Open Margin (no fence between the field and watercourse), Narrow Margin (< 2 m riparian buffers) or Wide Margin (> 4 m riparian buffers). At each site, two sampling transects were established, one at the water course (Water) and one 4 m into the field (Field). For Wide Margins a third transect was established in the middle of the buffer (Middle). This gave seven treatments: Open Water, Open Field, Narrow Water, Narrow Field, Wide Water, Wide Middle and Wide Field.

Along each transect, surface-active invertebrates were monitored by pitfall trapping, randomly placed quadrats (1 m x 1 m) were used to determine vegetation composition, and vegetation density was measured using a Robel pole (Robel *et al.*, 1970). Linear mixed models (REML) were fitted to test for effects of treatment, vegetation density and year (after adjusting for farm and location) on the abundance of key invertebrate groups (Fixed Effects = Treatment+VegetationDensity + Year; Random Effects = Farm/Location/Year).

## Results and discussion

With the exception of Lycosidae spiders, the activity abundance of all groups of invertebrates was significantly influenced by vegetation density and treatment (Table 1). Harvestmen (*Opiliones*), sawfly (*Symphyta*) larvae and *Cicadellidae* plant bugs had higher activity abundances in riparian buffers, but not in open riparian zones, when compared with the adjacent fields, indicating the exclusion of livestock from the margins favoured these invertebrates. Limacidae slugs also showed a similar trend, with the activity abundance of this group being greater in wide margins (but not narrow margins) than the adjacent fields. As harvestmen and slugs are prone to desiccation, the denser vegetation and deeper litter layer associated with the riparian buffers may provide a more humid and hence favourable micro-climate. Higher activity densities may also be related to food and it is likely that sawfly larvae and *Cicadellidae* plant bugs, being phytophagous, are favoured by the higher diversity of plants in the fenced riparian margins.

Table 1. Results of REML analyses on the activity abundance of key invertebrates showing Wald statistic (W) and probability value.

Invertebrate group	Vegetation density (df = 1)	Influence density	Treatment (df = 6)	Location of difference (W = Water, M = Middle, F = Field)
Limacidae	W = 7.72 P = 0.005	+ve	W = 19.37 P < 0.005	All Wide > OpenF WideM > WideW/F & NarrowF
Carabidae	W = 30.96 P < 0.001	-ve	19.36 P < 0.001	OpenW & AllF > Narrow/ WideW & WideM
<i>Erigone</i> spp.	W = 46.82 P < 0.001	+ve	W = 13.33 P < 0.001	AllF > Narrow/ WideW & WideM OpenF > OpenW > WideM > WideW
Lycosidae	W = 0.34 P = ns	-	W = 8.09 P = ns	-
Opiliones	W = 29.24 P < 0.001	-ve	W = 60.96 P < 0.001	NarrowW, WideW/M > AllF NarrowW > OpenW OpenW & WideF > NarrowF
Symphyta larvae	W = 23.12 P < 0.001	+ve	W = 19.60 P < 0.005	All W > Wide/ NarrowF NarrowW > OpenF; WideM > WideF
Cicadellidae	W = 38.27 P < 0.001	+ve	W = 49.09 P < 0.001	NarrowW & WideM > AllF & Open/ WideW

*Erigone* spiders and *Carabidae* responded differently to the previous groups with these predominantly predatory groups tending to have a higher activity density in the field and open water sites when compared to the riparian buffers. *Erigone* spiders and many species of *Carabidae* are highly mobile with a high fecundity, and are consequently better adapted to cope with the highly fluctuating habitats typical of intensive grasslands (Luff and Rushton, 1989).

*Carabidae* and harvestmen were the only groups that had a negative relationship with vegetation density, indicating their activity abundance was lower in denser vegetation. As harvestmen are prone to desiccation, they may be predicted to be more abundant in the higher humidity associated with denser vegetation. This was clearly not the case and it is possible that dense vegetation may impede hunting. Both harvestmen and *Carabidae* predominately

consist of predatory species, and sparser vegetation may facilitate the detection and capture of prey (Telfer *et al.*, 2000).

## Conclusions

The exclusion of livestock from riparian margins favoured several groups of invertebrates (e.g. sawfly larvae, *Cicadellidae* bugs and harvestmen) which are important dietary components for many farmland birds. Consequently the adoption of riparian buffers in intensively managed grassland has the potential to increase farmland biodiversity, both at the field level, by increasing the abundance of key invertebrates, and at the landscape level, by increasing habitat heterogeneity.

## Acknowledgements

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# **A simplified method to determine the abundance of grass functional groups in natural grasslands**

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## **Abstract**

A functional classification of grass species into four groups has been proposed, based on their leaf dry matter content. These plant functional groups (PFTs) show well differentiated properties for vegetation use (e.g. digestibility). The aim of this work was to assess the reliability of an easy methodology for identification of the dominant grass group in a given grassland community to facilitate technical advising. Three methods of botanical relevés were tested on grasslands of beef and dairy production systems of the Aubrac region (Central Massif) in France. Methods display differences in complexity of field work: a complete botanical description (Braun-Blanquet), the method of de Vries and de Boer, and a simplified non-destructive adaptation of the latter. Data analyses were performed using Spearman rank correlation tests on grassland classification according to different variables, mainly species richness and proportion of grass functional groups. Results show differences in the number of species listed. No differences were observed between grassland classifications based on PFTs dominance. We conclude that the simplest of them could be used to assess the grass dominant group necessary to implement grassland management tools.

Keywords: botanical composition, grass contribution, functional groups of grasses, grassland typology, grassland management

## **Introduction**

Agronomic advice on the management of permanent grassland has long been based on the floristic composition of the plant communities. Shortage of time or knowledge for the identification of species has hindered the widespread adoption of this approach in the daily work of advisers. Research work aimed at simplifying the diagnosis of the grassland flora has led to the use of a functional approach (Lavorel and Garnier, 2002). A functional classification of grass species into groups has been proposed by Ansquer *et al.* (2004) according to their leaf dry matter content. This has resulted in the establishment of four functional types of perennial grasses (A, B, C and D), characterized by their dry matter content of water saturated leaves (LDMC). These groups are also distinguished by their habitat preference (from fertile to less fertile), their phenology (from earlier to later earliness) and their digestibility at the vegetative stage (from more to less digestible). These functional types, established at the species level, in turn allow permanent grasslands to be categorized as A, B, C and D according to the dominant grasses in the community. The work of identifying species is thus reduced to simply identifying the dominant perennial grasses in each field. Nevertheless, this identification can be very time consuming if exhaustive methods are used. The objective of this work is to test a simplified method of determining dominant functional groups in a meadow, in contrast to methods normally used in botanical or ecological surveys.

## **Materials and methods**

We studied 60 mown or grazed permanent pastures (Aveyron, France) varying in the amount of applied fertilizer and in the types of cutting and grazing management. Three methods of botanical surveying were compared in order to record in each field the following variables:

total number of species, proportion of grasses, and proportion of functional types (A, B, C or D). Species of these groups have preference for high or low fertility conditions, which corresponds respectively to species having capture resource strategies (A and B) or conservation resource strategies (C and D).

The method of de Vries and de Boer was used on 60 fields where an exhaustive botanical survey was carried out using a procedure adapted from the frequency rank method (de Vries and de Boer, 1959). Sampling was done along a transect of 20 handfuls (10 x 10cm) spaced about 5 m apart, with thorough sorting of the different species and estimation of the fraction of biomass of each species (using a score [0-6] for the abundance of each species, the species present but scarce being scored 0). For a given field, the relative abundance score for each species was calculated as the sum of the scores obtained for each species in the various handfuls/the maximum score obtainable ( $20 \times 6 = 120$ ).

The second method (Simplified), used on the same 60 fields, was a quick simplified botanical survey (30 minutes per field), simply identifying the dominant species. The recordings were made along a transect in 10 equidistant 40 cm quadrats. In each quadrat, we recorded the visual abundance of the main species by using the same scoring system as in the De Vries and De Bauer method. Species with less than 15% abundance within the quadrat were not recorded. For each field, we therefore have a number of species which are the only dominant ones, together with the relative abundance of these dominant species.

The third method (Braun-Blanquet) was applied to a sub-sample of 13 fields (4 mown meadows, 5 valley pastures and 4 summer pastures) out of the ones used for the other two methods. This enabled us to draw up an exhaustive list of all the species present on a total area of 256 m<sup>2</sup> (a square 16 m x 16 m). An abundance-dominance score was given to each species by taking account of its fractional soil cover, using the abundance-dominance method of Braun-Blanquet (1928). The proportion of grasses and functional types was calculated from these abundance-dominance scores. Table 1 summarizes the characteristics of the three methods used and the variables obtained for each field.

Table 1. Characteristics of the records from the different methods and the variables obtained.

Method	Number of fields	Sampling	Species recorded	Area sampled	Average time	Number of species	Abundance of species
De Vries & De Boer	60	100m transect 20 handfuls 10x10 cm	All	0.2 m <sup>2</sup>	4 h (2 people)	Yes	Yes
Simplified	60	Field diagonal 10 quadrats 40x40 cm	Dominant species	1.6 m <sup>2</sup>	30 min. (1 person)	No	On dominant species
Braun-Blanquet	13	Percentage ground cover	All	256 m <sup>2</sup>	4 h (2 people)	Yes	Yes

## Results and discussion

The data obtained enable one to compare the three methods over 13 fields, and the methods of De Vries and De Bauer and the Simplified method over all 60 fields. Table 2 shows the correlation coefficients obtained for the three methods for two of the variables studied on the 13 common fields. The third variable (N), which was not determined directly in the Simplified method, does not show a significant correlation with the other two. The percentage of grasses obtained by the Simplified method is significantly correlated with the values obtained by the other two methods. On the other hand, the percentages of grasses obtained by methods of Braun-Blanquet and de Vries and de Boer are not significantly correlated because

of the difference in the methods for estimating the proportions of species, which was based on biomass in the case of the de Vries and de Boer method, and on ground cover in the case of Braun-Blanquet. The percentages of grass types A + B (Species having resource capture strategies) are strongly correlated between the methods of Braun-Blanquet and de Vries and de Boer, and also with the Simplified method.

Table 2. Correlation coefficients (Pearson) for the variables proportion of grasses (% grasses) and percentage of functional types having a resource capture strategy (% A + % B) between the methods compared in pairs (n = 13).

Variables	Method		
	De Vries & De Boer and Braun-Blanquet	De Vries & De Boer and Simplified	Braun-Blanquet and Simplified
% grasses	0.53 <sup>ns</sup>	0.68 *	0.79 ***
%A + %B	0.87 ***	0.93 ***	0.84 ***

\*\*\*  $P \leq 0.001$ , \*  $P \leq 0.01$ , ns Not significant.

Table 3 summarizes the Spearman correlation coefficients (data not normally distributed) for the two methods used on 60 fields (De Vries and De Bauer and Simplified) on the grass variables of type A + B and of each functional type considered separately. It is clear that the classifications of the fields are very significantly correlated for all the variables considered.

Table 3. Correlation coefficients (Spearman) for the variables proportion of grasses (% grasses) and proportions of functional types, A (%A), B (%B), C (%C) and D (%D) between methods of De Vries and De Bauer and Simplified (n = 60).

	% Grasses	% A	% B	% C	% D
$r^2$	0.54 ***	0.83 ***	0.82 ***	0.87 ***	0.46 ***

\*\*\*  $P \leq 0.001$ .

## Conclusions

All these results therefore show a close agreement between the proportions of grasses and of functional types observed using the Simplified method and methods using exhaustive botanical data, used on 13 (De Vries and De Bauer and Braun-Blanquet) or 60 (De Vries and De Bauer) fields. The quick simplified recording method (Simplified) may therefore be used by agricultural advisers to characterize grasslands, since it enables one to detect the grass functional groups forming the majority of the grassland biomass. Furthermore, it only requires a short working time in the field (30 minutes) and simplifies the botanical expertise to the recognition of the dominant grasses.

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# Specific and functional diversity of vegetation in three beef cattle farms of the French Massif Central

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## Abstract

The aim of this pilot study was to analyse the specific and functional diversity of vegetation on three beef cattle farms taken within a stocking rate gradient, as well as the range of between-plot diversity in each farm. The farms were located in the same upland area with similar altitude, rainfall and soil conditions, and they relied exclusively on semi-natural grasslands. Average stocking rates were 1.2, 1.0 and 0.7 LSU ha<sup>-1</sup> in F1, F2 and F3. Botanical surveys were carried out per vegetation facies on 9-11 representative plots in each farm. An average number of 37, 55 and 68 plant species per plot was recorded in F1, F2 and F3 ( $P < 0.001$ ), which suggests an increase in plant diversity as stocking rate decreases. The total number of species measured was 123, 144 and 194 in F1, F2 and F3, respectively. The widest range of between-plot diversity was, however, recorded on the farm with an intermediate stocking rate. The relative abundance of grass functional types was explained by differences in grazing management. Higher species richness in grazed compared with cut plots was due to the presence of more vegetation facies and to a lower nitrogen fertilization.

Keywords: biodiversity, functional traits, grasslands, farm level, stocking rate

## Introduction

Improved grassland biodiversity has frequently been associated with extensive farming systems with a reduction of fertilization inputs and stocking rate. Additionally, the diversity of management practices (cutting vs. grazing, stocking rates, etc.) at the farm scale could also increase biodiversity as the result of the creation of different habitats for plant and animal communities. The first aim of this pilot study was to describe the level of plant diversity in three beef cattle farms of the French Massif Central, which were taken within a stocking rate gradient. We then analysed between-plot diversity in each farm, and related it to differences in plot management.

## Materials and methods

The study was conducted on three beef cattle farms (F1-3) in the upland area of French Massif Central (Farruggia *et al.*, 2006), which rely exclusively on semi-natural grasslands. Farm size was 115, 119 and 63 ha, with herd sizes of 100, 74 and 38 Salers cows in F1, F2 and F3, respectively. This corresponds to average stocking rates of 1.2 (F1), 1.0 (F2) and 0.7 (F3) LSU ha<sup>-1</sup> (with 1 LSU = 1 cow), representative of those that can be found in this type of farming system. Percentage of areas for cutting ranged from 41 to 61% of the total farm area. Average mineral nitrogen fertilization decreased from 60 to 10 kg ha<sup>-1</sup> between F1 and F3. F1 was also characterized by the use of cut areas for silage and by strip grazing, whereas in farms F2 and F3 cut areas were hay meadows, and pastures were continuously or rotationally grazed. The three farms were located in a 30-km radius circle in an area that is homogeneous for edaphic conditions (volcanic soils), altitude (from 1000 to 1300 m) and rainfall (> 1000 mm yr<sup>-1</sup>). Global surveys were first carried out on the farms to understand management rules and to make an inventory of the different plots, in order to select between 9

and 11 representative plots per farm (half of which were cut) to carry out botanical measurements. In each plot, percentage cover of all plant species was estimated visually on a 16-m long transect per vegetation facies (if facies area > 15% of plot area), with one point every 50 cm. Any additional species occurring within 5 m of the transect were also recorded. A total number of 54 transects within 31 plots was thus described, i.e. 14, 19 and 21 transects in F1, F2 and F3, respectively. We used the total number of plant species per plot (tN) as a first biodiversity indicator. The average balanced number of plant species per plot (bN) was then calculated from the sum for the different facies of the number of plant species per facies multiplied by facies area (in % of plot area). Grass species were also classified into four different functional types according to the classification, based on vegetative (mainly leaf DM content) and reproductive traits, proposed by Cruz *et al.* (2002). These types are also associated with different defoliation regimes and fertilization inputs (Table 1). Balanced percentage of each functional type (bA, bB, bC and bD) were calculated per plot in the same way than for bN.

Table 1. Functional groups for grass species according to Cruz *et al.* (2002).

	Rich/fertile swards	Poor swards
Adapted to frequent defoliation regime	<b>Type A:</b> high specific leaf area (SLA), high digestibility, short leaf lifespan, early reproductive growth	<b>Type C:</b> low SLA, medium digestibility, long leaf lifespan, medium-to-late reproductive growth
Adapted to lenient defoliation regimes	<b>Type B:</b> medium SLA, high digestibility, long leaf lifespan, medium-to-late reproductive growth	<b>Type D:</b> low SLA, low digestibility, very long leaf lifespan, late reproductive growth

To characterize vegetation diversity at the farm scale, we also used the total number of plant species recorded on a farm, and the average number of facies per plot in grazed and cut plots (Farruggia *et al.*, 2006). Finally, we classified plots according to their level of specific diversity, conventionally considered as low (L) for bN < 30, medium (M) for bN between 30 and 40, and high (H) for bN > 40, which allows a first estimate of between-plot diversity.

At the plot scale, we used the GLM procedure of SAS to analyse differences in species numbers, grass functional types and facies numbers between farms and management practices (cut vs. grazed plots). Differences between treatments were detected using the Tukey-Kramer correction for multiple comparisons.

## Results and discussion

A total number of 123, 144 and 194 species was recorded in F1, F2 and F3, respectively. Both the number of plant species (tN and bN) and the number of facies per plot significantly increased between F1 and F3 (Table 2).

Table 2: Plant diversity and functional composition of swards in plots of the three farms.

	F1	F2	F3	s.e.	P
tN	37 <i>a</i>	55 <i>ab</i>	68 <i>b</i>	19	***
bN	30a	35a	45b	5	***
Number of facies per plot	1.3a	2.3b	2.2b	0.9	*
bA (%)	27	19	10	16	t
bB (%)	26	25	19	14	N.S.
bC (%)	38	43	56	20	t
bD (%)	2 <i>a</i>	7 <i>ab</i>	11 <i>b</i>	8	***

For each line, means with different superscripts differ: \*\*\*  $P < 0.001$ ; \*\*  $P < 0.01$ ; \*  $P < 0.05$ ; t:  $P < 0.10$ .

The relative abundance of competitive type-A species also tended to be higher in F1 plots, whereas conservative types C and D were more abundant in F3 plots, which is consistent with differences in average fertilization and defoliation regimes between the three farms.

Independently of the farms, plot management had a great impact on sward diversity since tN and bN were significantly higher in grazed compared with cut plots (68 vs. 46:  $P < 0.001$ ; 40 vs. 35:  $P < 0.10$ , respectively). This can be explained by the greater heterogeneity of grazed pastures (2.3 vs. 1.5 facies per plot in cut swards,  $P < 0.05$ ) combined with their lower level of fertilization. Indeed, mineral nitrogen supply reached an average of 15 kg N ha<sup>-1</sup> for grazed plots vs. 25 kg N ha<sup>-1</sup> for cut plots, with additional organic fertilization for the cut plots. Consistently, grasses from types C (51 vs. 42 in grazed vs. cut plots,  $P < 0.05$ ) and D (16 vs. 3,  $P < 0.001$ ) were favoured in the grazed plots, whereas the opposite was found for type-B grasses (13 vs. 30,  $P < 0.001$ ). Similar differences have been reported by Duru *et al.* (2005) in the French Pyrenees. Finally, the widest range of between-plot diversity was recorded on farm F2, where plots with the three levels of diversity were present (Figure 1).

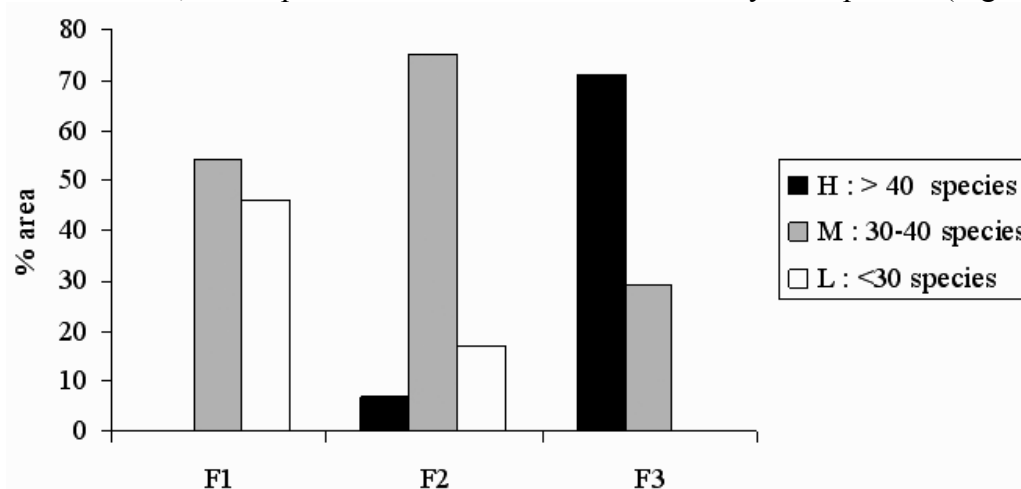


Figure 1: Range of between-plot plant diversity in the three farms.

## Conclusions

This pilot study illustrates the relationships between management rules and the specific and functional diversity of vegetation at the farm and the plot scales. In these three farms, which were taken within a stocking rate gradient representative of those that can be found in beef farming systems of the French Massif Central uplands, the greatest specific diversity was reached when fertilization input and defoliation intensity were the lowest. Besides that, the range of between-plot diversity differed between farms. We also underline the lower level of diversity in cut compared with grazed plots. In this farming system, cut plots are chosen for their topography in order to facilitate silage and hay cuts and receive higher fertilization inputs, all these factors increasing plot homogeneity and reducing plant diversity. Further studies are now conducted within a larger farm network, with an emphasis on the effect of between-plot diversity on overall farm biodiversity and forage autonomy.

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# Effect of intensifying land use on community structure, functional diversity and productivity in calcareous grasslands

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## Abstract

This study aims at clarifying the impact of increasing land use on specific and functional structure of herbaceous plant communities and how they influence biomass production. The study was conducted at the 'La Fage' experimental station, located on a limestone plateau in southern France. Three land use treatments, corresponding to a gradient in intensity of use, were applied on these rangelands: sheep grazing since 1972, fertilization of grazed parcels since 1978, and abandonment since 1987. Botanical records and biomass harvests were regularly performed in these treatments. Traits of species constituting 80% of the standing biomass were measured in 2006-2007. Both the composition of communities and their functional structure strongly depend on land use treatment, and are highly correlated with biomass production. The reconstruction of communities' functional trajectory supports the predictive power of the 'functional trait approach'. The mechanistic link between key plant traits related to plant growth and ecosystem productivity demonstrates the relevance of this method to predict the outcomes of future land use modifications.

Keywords: community structure, functional trait, grassland productivity

## Introduction

Land use changes are important drivers of global change (Costanza *et al.*, 1997; Chapin *et al.*, 2000). Understanding and predicting plant community responses, ecosystem properties and their associated services are therefore major objectives in both applied and theoretical ecology (Hooper *et al.*, 2005). To face this challenge and to avoid the strong limitations of taxonomy-based classifications, the functional trait approach was developed. Analyses of community response to grassland management (grazing and its abandonment) demonstrated the relevance of key plant traits (see Diaz *et al.*, 2007 for a review). Intensive grassland management favours plants with rapid resource acquisition (high specific leaf area, low tissue density, and high leaf nitrogen content). Abandonment creates the opposite response, increasing the dominance of conservative strategies.

In this paper we examine i) the response of herbaceous plant communities in terms of specific and functional structure to the intensification of land use, and ii) the influence of such changes in community composition on ecosystem functioning – biomass production.

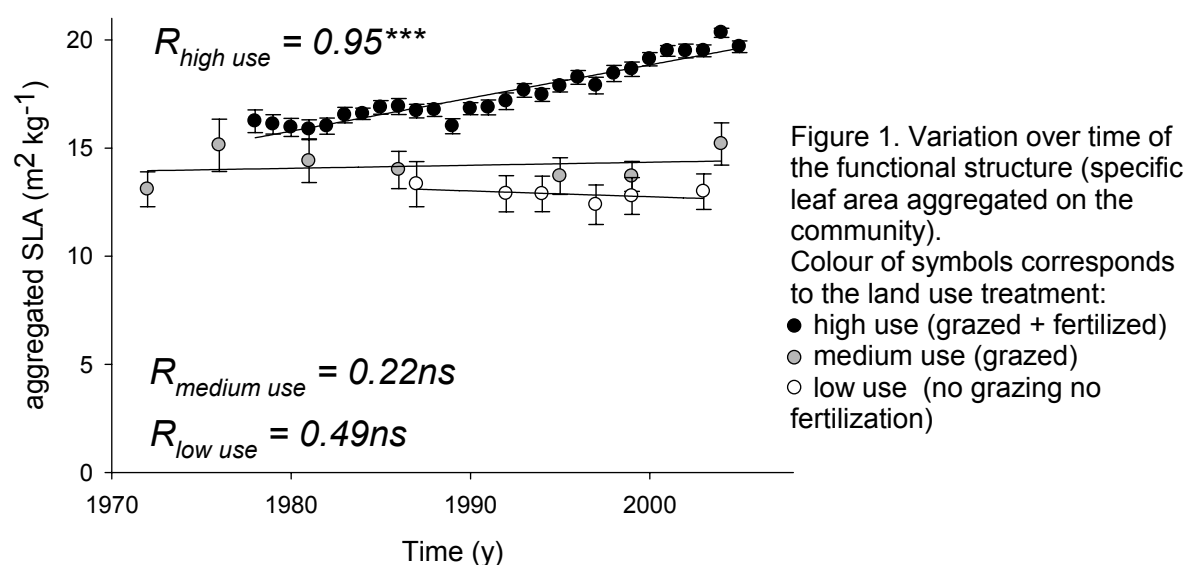
## Materials and methods

The La Fage experimental station is located on the Larzac Causse, a limestone plateau in southern France. The site has been sheep grazed (medium-use treatment) since 1972. Mineral

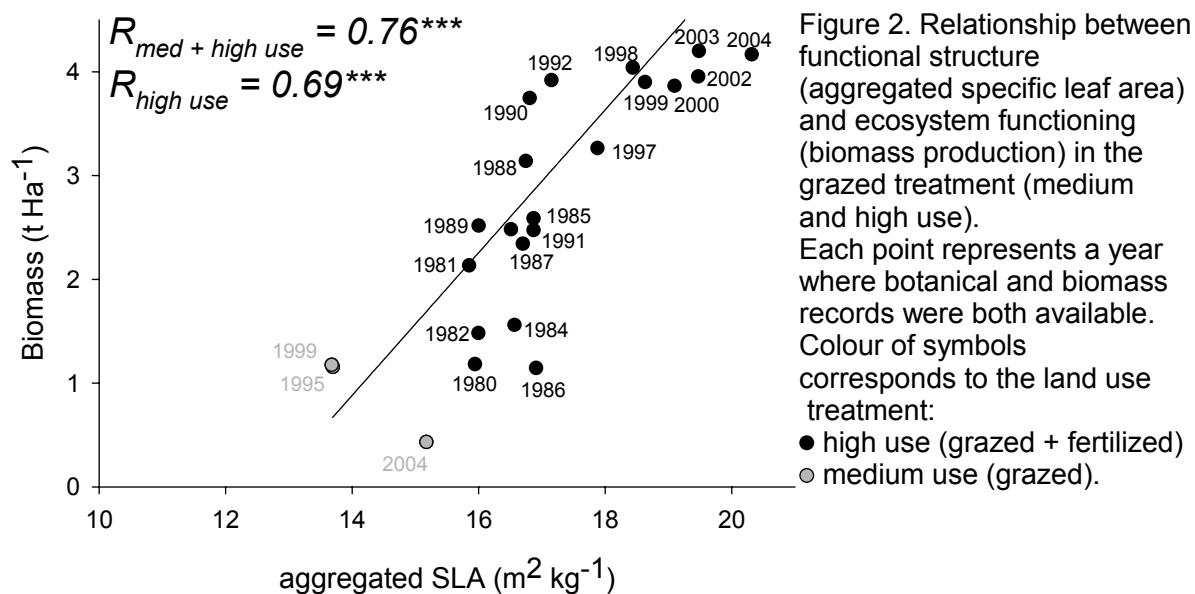
fertilization (NP) has been applied on two parcels (high use treatment) since 1978. A low-use treatment (ungrazed and unfertilized) was created in 1987. Botanical records were conducted every year in the high-use treatment and every five years elsewhere, using point-quadrat survey lines. Species life cycle was extracted from local flora to obtain the representation of annual species on each botanical record. Biomass was harvested every year since 1990 and 1980, respectively, in the medium- and high-use treatments. Specific leaf area (SLA), ratio of leaf projected area to its dry mass, was screened in 2006-2007 on the dominant species (contributing to 80% of biomass). The community functional parameter (aggregated SLA) was calculated as the mean SLA value per species weighted by species relative abundance (Violle *et al.*, 2007).

## Results and discussion

Fertilization associated with grazing induces a significant variation of community structure. In the high-use treatment, rangelands initially composed of herbaceous perennials significantly shifted to mixed communities, with the proportion of annuals increasing from 1% to 50% in 30 years. Despite changes in species dominance with time, the proportions of life cycle remained stable in the low- and medium-use treatments with 1% to 5% of annuals.



Changes in community structure in the high-use treatment are strongly associated to functional changes (Figure 1), due to the increasing abundance of fast growing species characterized by high SLA values. No significant change is recorded in the other low- and medium-use treatments, where slow-growing conservative species dominate. Slow growing species replacement by fast growing species with increasing land-use corresponds to an increase in biomass production (multiplied by 5 since the beginning of fertilization). The strong dependence of biomass production on aggregated specific leaf area (Figure 2) confirms that plant traits related to resource use strategy can be used to scale up from organ to ecosystem functioning (biomass production) in complex plant communities (Garnier *et al.*, 2004).



## Conclusions

The intensification of land use induces large changes in the composition and functional structure of communities, resulting in changes in biomass production, superimposed on the direct effect of fertilizer addition. This study confirms that leaf traits involved in the acquisition–conservation trade-off such as specific leaf area are useful tools for the analysis of community responses to land use changes. This temporal reconstruction of communities' functional trajectory supports the predictive power of the 'functional trait approach'.

## Acknowledgements

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# Tree density and fertilization regime effect on pasture biodiversity in a silvopastoral system established under *Pinus radiata* D. Don

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## Abstract

Land use change, such as afforestation, has been one of the most important goals achieved in the last years at a European level. Promotion of afforestation and agroforestry systems can later affect biodiversity in either a positive or negative way. The objective of this work is to evaluate the effect of two different tree densities (established at 2500 and 833 trees ha<sup>-1</sup>) of *Pinus radiata* D. Don, and three types of fertilization (no fertilization, inorganic fertilization and organic fertilization) on alpha (Simpson index) and beta (Jaccard index) biodiversity. Canopy development reduced biodiversity as there was no seed bank with shade-adapted species present. When no richness biodiversity parameter was affected, the relative composition under each kind of agroforestry system was clearly modified over time.

Keywords: agroforestry, Simpson, Jaccard

## Introduction

Agroforestry systems are enhanced and promoted by the recent European Union (EU) Rural Programme (UE 2006). Agrarian policy measures for afforestation have an important impact in Galicia (NW Spain), and in Europe generally, where more than one million hectares were afforested in the last decade (Rois *et al.*, 2006). EU policies also seek to reduce biodiversity losses by 2010, land use change can cause important modifications to biodiversity.

Biodiversity is an important issue to promote agricultural sustainability, and depends on vegetation management. Biodiversity in extensive systems can enhance productivity due to the complementarity of different plant species in their use of soil resources, i.e. cover-grass association. Fertilization usually reduces biodiversity due to the enhancement of a few, very competitive, species. Complex interactions between fertilization effects and agroforestry systems can be found due to the temporal modification of microclimatic conditions as tree cover is developed. This aspect is especially relevant when fast-growing species are used in areas with high tree growth potential, as in Galicia. Biodiversity studies are mainly based on richness, but also evenness is important to enhance biodiversity benefits at a high level.

## Materials and methods

The experiment was established in spring 1995 in the Atlantic Spanish area of Lugo (NW Spain) following a randomized-block design. *Pinus radiata* D. Don afforestation was made at two densities (2500 and 833 trees ha<sup>-1</sup>) on an area of abandoned agricultural land that had an initial soil pH of 6.8. Every plot consisted of 25 trees distributed in a square of 5 x 5 trees. Pasture was established after soil preparation with two seeds mixtures, Dg and Lp. Dg consisted of: 25 kg ha<sup>-1</sup> *Dactylis glomerata* L. var Saborto, + 4 kg ha<sup>-1</sup> *Trifolium repens* L. Var Ladino, + 1 kg ha<sup>-1</sup> *Trifolium pratense* L. var Marino, and mixture Lp consisted of 25 kg ha<sup>-1</sup> *Lolium perenne* L. var Brigantia, + 4 kg ha<sup>-1</sup> *Trifolium repens* L. var Ladino, + 1 kg ha<sup>-1</sup> *Trifolium pratense* L. var Marino. Three fertilization treatments were applied: NF (nil-fertilization), L (dairy sewage sludge fertilization at a rate of 154 m<sup>3</sup> ha<sup>-1</sup> at the establishment), and M (500 kg 8:24:16 at the beginning of every year, plus 40 kg N ha<sup>-1</sup> after first

harvest). Treatment L received the same treatment as M after 1997. Every year pasture production was estimated by harvesting an area from between six inner trees in the plots, three times during the spring and once in the autumn, with the exception of the first year, when two harvests, one per season, were made. Samples were transported to the laboratory, where species were hand separated and botanical composition was estimated based on percentage of dry matter. Alpha (Simpson index  $((1-\lambda))$  and beta biodiversity (Jaccard Index ( $I_j$ )) of the first, fifth and tenth year of the experiment were estimated (Magurran, 1989). ANOVA and Duncan test was used for statistical analyses.

## Results and discussion

The Simpson index represents the probability that two randomly selected individuals in the habitat belong to the same species. Its value is in inverse equity therefore, diversity can be calculated as  $(1-\lambda)$  (Lande, 1996). The index varies between 0 and 1, with the lowest values to the situations in which the system presents a minimal relative diversity (Magurran, 1989; Moreno, 2001). At 2500 trees  $\text{ha}^{-1}$  the Simpson index varied between 0.71-0.80, 0.60-0.84 and 0.00-0.31 and at 833 trees  $\text{ha}^{-1}$  0.58-0.79, 0.50-0.77 and 0.48-0.75 for 1995, 2000 and 2005 (Figure 1). Lowest Simpson index values, and therefore lowest biodiversity, were found in treatments with high canopy development (high density stand: 2500 trees  $\text{ha}^{-1}$ ). Tree development in the last year made the microhabitat more homogeneous, and in this situation, only those species adapted to shading can survive, and the increase in canopy cover caused a reduction of biodiversity compared with the open areas (Díez *et al.*, 1992; Ferrer-Benimeli, 1999). The development of the different agroforestry systems modified soil and pasture characteristics as was described for this experiment by Mosquera-Losada *et al.* (2006). Sludge and no fertilization treatments had higher tree development than mineral fertilization, which caused changes in biodiversity.

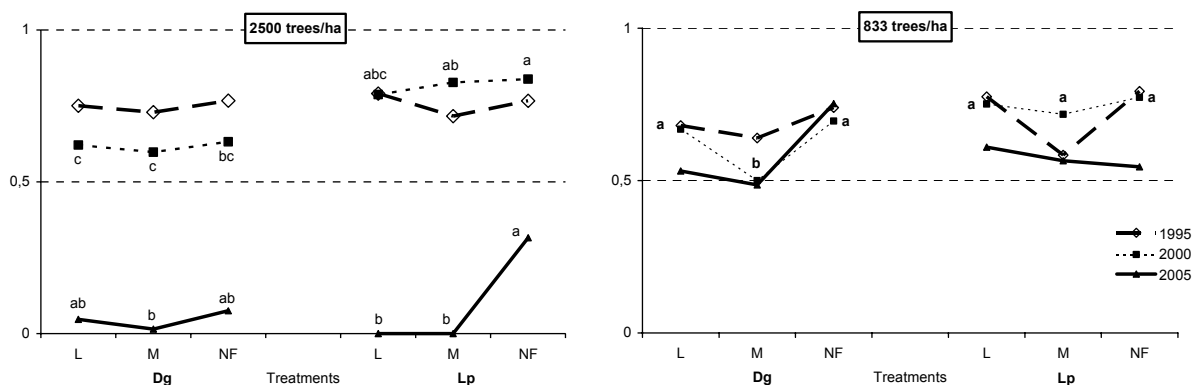


Figure 1. Simpson index  $(1-\lambda)$  for three years and two density where: L: dairy sewage sludge fertilization; M: inorganic fertilization; NF: None fertilization; Dg: cocksfoot mixture and Lp: ryegrass mixture.

Beta biodiversity, described as Jaccard index ( $I_j$ ) can be seen in Table 1. This index enables us to compare two different land uses, agricultural land and forest land, since the change in land use causes changes in the floristic diversity (Gustavsson *et al.*, 2007). As was found with the Simpson index, it has been found that the canopy increment reduced a number of species in time, probably due to the tree-pasture competition (Brewer, 1998), but also the lack of the pasture species adapted to showing in previous open pastureland. However, even though the number of species is not affected when the tree cover parameter is low, the composition is very different. It has been found that only 35% of the species developed under are the same during the ten years of study. Burel *et al.* (1998) has indicated that intensification could not



modify richness, but the species composition could not be the same. This modification is mainly explained by the annual-perennial species relationship in the first and last year of this study.

Table 1. Jaccard Index ( $I_j$ ) for three years and two density where: a = species in the parcels at the beginning of trial (year 1995); b = species in the parcels at the end of trial (year 2005) and c = number of species present in both situations.

Density	Jaccard Index ( $I_j$ )		c	$I_j$
	a <sub>95</sub>	b <sub>05</sub>		
2500 trees ha <sup>-1</sup>	34	9	4	0.10
833 trees ha <sup>-1</sup>	31	31	17	0.38

## Conclusions

Biodiversity can be modified not only by the change in land use but also by the evolution of the grassland since it was sown. Land use change which modifies ecological factors (light and indirectly temperature) and soil characteristics should be taking into account to reduce biodiversity losses. Tree cover development, enhanced by initial sewage sludge fertilization, and the nil-fertilization treatments reduced significantly the biodiversity (as Simpson index). Beta-biodiversity changed also with time, mainly due to the annual-perennial species relationship.

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# Diet selection and herbage intake by cattle and sheep grazing two contrasting heathland communities

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## Abstract

Heather moorland is recognized as having international importance, yet there has been a decline in the extent and condition of this habitat as a result of inappropriate management. In this experiment the diet composition and herbage intake of cattle and sheep grazing heathland swards with Low (8%) and High (61%) percentages of cover of *Calluna vulgaris* were estimated from faeces profiles of *n*-alkanes and long-chain fatty alcohols. Two breeds of sheep (Welsh Mountain, Scottish Blackface) and two breeds of cattle (Welsh Black, Continental cross) grazed each experimental site during two separate sampling sessions in July and September. On each site both cattle and sheep were selective feeders, consuming grasses in preference to dwarf shrub. The quantities of *C. vulgaris* consumed were small, even on the High site, reflecting the biomass of the graminoid species present, and hence availability of preferred items. There were several significant differences in the proportions of different plant groups consumed by the cattle and sheep. There were also significant differences in the composition of the diets selected by the different breeds of sheep, whereas the diets of the two cattle breeds were generally similar.

Keywords: cattle, sheep, breed, *Calluna vulgaris*, diet composition, herbage intake

## Introduction

Heather moorland is recognized as having international importance from a nature conservation perspective, yet there has been a decline in the extent and condition of this habitat in the UK as a result of inappropriate management. In particular, heavy grazing by sheep and other large herbivores has been linked to a reduction in *Calluna vulgaris* and similar dwarf shrub heath species, and their replacement by graminoids. In the UK the funding mechanism for moorland restoration is being implemented primarily through agri-environment schemes, yet to date remarkably few comparative grazing studies with domesticated livestock have been conducted on this type of vegetation community. Clarifying the relationship between sward characteristics and the grazing behaviour of different types of livestock grazing moorland is vital if appropriate management guidelines are to be produced. The objective of this study was to investigate the interactions between animal type, sward composition and diet selection in order to improve understanding of factors influencing the impact of grazing on heather moorland.

## Materials and methods

Separate experimental sessions were conducted on heathland swards with Low (8%) and High (61%) percentages of cover of *C. vulgaris*. Two breeds of sheep (Welsh Mountain (WM), Scottish Blackface (SBF)) and two breeds of cattle (Welsh Black (WB), Continental cross (CX)) (n = 6 per breed) grazed each experimental site during two separate 14-day sampling sessions, in July and September respectively. The experimental design adopted followed that of earlier studies of comparative grazing on similar swards (Grant *et al.*, 1987). To ensure the

presence of one type of animal did not influence the behaviour of another, a 4 ha measurement area at each site was divided into four subplots. The different species/breed groups rotationally grazed these subplots, moving on to the next subplot daily. In this way, all four animal types separately grazed the same measurement area over the course of each measurement week at each site.

At both sites the measurement area was characterized in detail in terms of sward composition and structure prior to grazing. Separate samples of the main plant species growing within each experimental site were taken for subsequent chemical analysis to determine chemical composition. Diet composition was estimated using *n*-alkane and long-chain fatty alcohol profiling, and feed intake and diet digestibility determined by dosing with *n*-alkanes for 12 days and collecting faecal grab samples for 4 days. Samples were bulked on an individual animal basis. Faeces concentrations of *n*-alkanes and long-chain fatty alcohols from animals at each site and each sampling session were subjected to principal components analysis using procedures in the Matlab (version 7.2) statistics toolbox to investigate data structure. For dietary components analysis of variance was carried out using orthogonal contrasts (Genstat 10.1). Diet composition data were subject to an angular transformation prior to analysis. Selectivity indices were calculated according to Jacob (1974).

## Results and discussion

Principal components analysis of faecal concentrations of *n*-alkanes and fatty alcohols showed separation between cattle and sheep, and between breeds of cattle and sheep, at each sampling time (as illustrated in Figure 1).

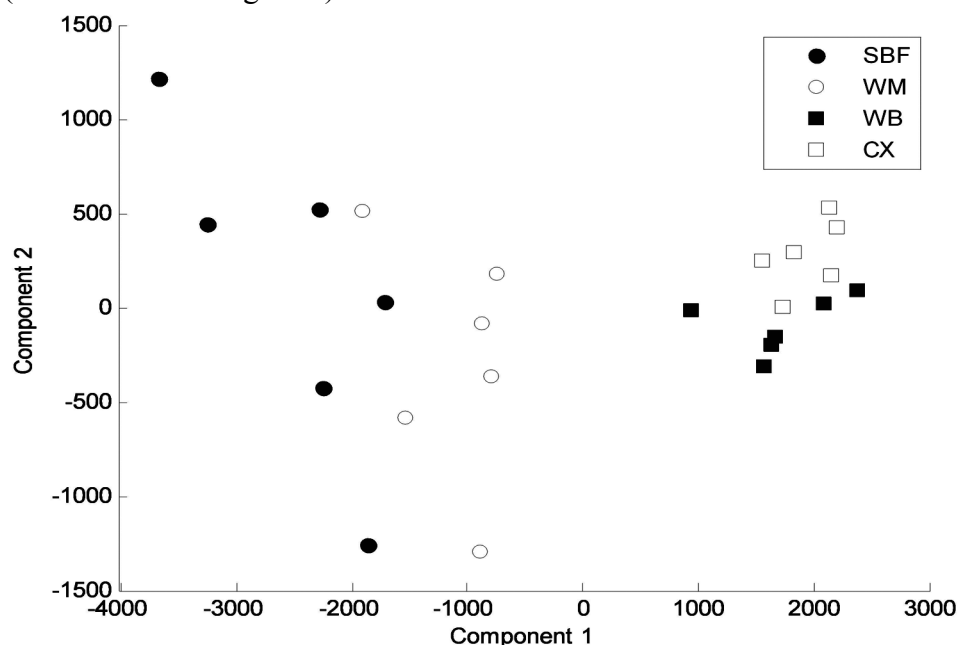


Figure 1. Principal components 1 versus 2 of faecal concentrations of *n*-alkanes and long-chain fatty alcohols sampled in September from animals grazing the High site (SBF = Scottish Blackface, WM = Welsh Mountain, WB = Welsh Black and CX = Continental cross).

At the Low site the diets of both animal species were dominated by grasses. In July the sheep diets were found to contain a significantly higher proportion of *C. vulgaris* ( $P < 0.01$ ) than the cattle diets, although overall this was a minor dietary component and accounted for  $< 3.5\%$  of the material consumed. Compared with the cattle diet, the sheep diet in September again contained more *C. vulgaris* ( $P < 0.001$ ). When the diets of the WM and SBF were compared there were significant between-breed differences in consumption of the majority of dietary

categories during both experimental sessions. These included *C. vulgaris* ( $P < 0.01$ ) and other dwarf shrub species ( $P < 0.05$ ). Selectivity indices revealed there had been consistent selection for grasses by both cattle and sheep during both sessions. Although *C. vulgaris* had been avoided by all groups, the strength of avoidance was less for the SBF sheep. Other dwarf shrubs were also avoided, but to varying degrees.

At the High site in July several between-species differences in diet composition were recorded. The sheep diets again contained more *C. vulgaris* ( $P < 0.05$ ), but the quantities consumed were still small. Previous studies have highlighted the relationship between availability of preferred food items and consumption of *C. vulgaris*, and thus the results obtained can be linked to the biomass of graminoid species present. There were no between-breed differences recorded for the sheep at this time, and the only difference between the cattle breeds was in the proportion of other dwarf shrubs ( $P < 0.05$ ). In September the sheep diets still contained more *C. vulgaris* ( $P < 0.001$ ). While the diets of the two breeds of cattle were similar at this time, there were a number of significant between-breed differences in the sheep diets, with the SBF diet containing substantially more *C. vulgaris* ( $P < 0.001$ ). When selectivity indices were calculated there was consistent selection for broad-leaved grasses during both sessions at this site. *Nardus stricta* was tolerated by the sheep and selected by the cattle in July, confirming earlier observations that cattle are less selective feeders than sheep, and will consume greater quantities of this invasive grass species (Grant *et al.*, 1987).

During both sampling sessions at the Low site the dry matter (DM) intake per unit metabolic liveweight of the cattle and sheep was similar. However, the DM digestibility (DMD) of the diet selected by the sheep was significantly higher on both occasions ( $P < 0.05$ ). In contrast, the differences in diet composition between the WM and SBF sheep were not reflected in between-breed differences in intake at this site. When grazing the High heather site in July there was no significant difference in the intake per unit live weight of the cattle and sheep, or in the digestibility of the diet. However, in September the sheep had a higher relative intake ( $P < 0.05$ ) and the diet consumed had a higher DMD ( $P < 0.001$ ). Between-breed differences in intake and diet DMD were recorded for both species during July ( $P < 0.05$ ).

## Conclusions

Although both cattle and sheep were confirmed to be selective feeders consuming grasses in preference to dwarf shrubs, key differences in diet composition were recorded, notably with regard to *C. vulgaris* and *N. stricta*, which have implications for the long-term impact of grazing by the two species. Similarly, the results from this study indicate that substantial differences exist in the foraging strategy of different breeds of sheep, and that quantifying these for a range of breeds may offer potential for land managers to meet environmental goals more effectively without compromising returns from conventional production systems. In contrast, the overall diet composition of different breed types of cattle was found to be similar. This implies that commercial breeds may have the potential to deliver many of the environmental benefits of grazing associated with traditional breeds.

## Acknowledgements

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# Performance of Limousin cross and Belted Galloway suckler cows and calves when grazing improved and semi-natural pasture

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## Abstract

A four-year systems experiment is quantifying the economic and environmental impacts of a) mixed grazing of sheep with cattle, b) removal of cattle from mixed systems on improved pasture to graze semi-natural rough grazing (SNRG) during the summer months, and c) grazing with a traditional rather than a modern breed of cattle. This paper reports initial findings with regards to cattle performance. From early June through to August/September spring-calving suckler cows were managed according to three treatments: 1) Limousin-cross cows and their Limousin calves mixed with sheep grazing *Lolium perenne*/*Trifolium repens*-dominant improved permanent pasture (Lim PP); 2) Limousin-cross cows with Limousin calves grazing *Molinia*-dominant SNRG (Lim SNRG); 3) Belted Galloway cows with Belted Galloway calves grazing *Molinia*-dominant SNRG (BG SNRG). Replicate plots were grazed by 4 cows and their calves plus 24 ewes and 36 lambs (Lim PP) or 4 cows and their calves (Lim SNRG; BG SNRG). Cattle were removed from the SNRG when the utilization of the current season's growth of *Molinia caerulea* had reached an average of 50%. In 2005 calf liveweight gain was higher for Lim PP than for Lim SNRG, and higher for BG SNRG than for Lim SNRG when differences in initial liveweight were taken into account. At this time the cows on all three treatments gained weight. In contrast, during 2006 the performance of the cows was markedly different with the Lim SNRG cows losing weight while the BG SNRG cows gained more weight than the Lim PP cows. During this grazing period liveweight gains by the calves of the two breed types on the SNRG were similar when expressed on a metabolic liveweight basis, but again these were lower than that of the calves on the Lim PP.

Keywords: liveweight gain, cattle, permanent pasture, rough grazing, breeds, native

## Introduction

Despite growing interest in the environmental benefits of grazing by cattle, and in particular the potential role of grazing in controlling invasive hill grass species such as *Molinia caerulea* within semi-natural plant communities, very little is known about the levels of cattle performance that can be achieved on this type of rough grazing. Previous research suggests that there are important differences in the way in which breeds of beef cow and their suckling calves perform in different nutritional environments. It is possible that the poorer nutritional environments in the uplands may penalize large continental breeds with high nutrient requirements more than traditional, slower growing breeds, but this has not been tested. In order to provide a scientific basis for the development of management guidelines for Less Favoured Areas a four-year systems experiment is quantifying the economic and environmental impacts of: a) mixed grazing of sheep with cattle, b) removal of cattle from mixed systems on improved pasture to graze semi-natural vegetation during the summer months, and c) grazing with a traditional rather than a modern breed of cattle. This paper reports on cattle performance during the first two years of data collection.

## Materials and methods

From approximately June to August spring-calving suckler cows are managed according to three treatments: 1) Limousin-cross cows and their Limousin calves mixed with sheep grazing *Lolium perenne*/*Trifolium repens*-dominant improved permanent pasture (Lim PP); 2) Limousin-cross cows with Limousin calves grazing *Molinia*-dominant semi-natural rough grazing (SNRG) (Lim SNRG); 3) Belted Galloway cows with Belted Galloway calves grazing *Molinia*-dominant SNRG (BG SNRG). Replicate plots were each grazed by 4 cows and their calves plus 24 ewes and 36 lambs (Lim PP) or 4 cows and their calves (Lim SNRG; BG SNRG), with plots sizes of 2.25 ha and 4 ha on the improved pasture and SNRG respectively. The improved permanent pastures (PP) had been reseeded at least 10 years previously. They were dominated by perennial ryegrass (*Lolium perenne*) with a white clover (*Trifolium repens*) content of approximately 5-10%. Unsown grasses were mainly bent (*Agrostis* spp.), meadow grasses (*Poa* spp.) and Yorkshire fog (*Holcus lanatus*). Four of the areas of *Molinia caerulea*-dominated SNRG grazed during the experiment had been entered into an agri-environment scheme in 1996 as species-rich pastures.

Cattle were turned out onto the SNRG when there was sufficient new season's biomass to support them. To monitor utilization of *Molinia caerulea* six enclosure areas (2 m x 2 m) were erected on each SNRG plot at the start of the growing season. At fortnightly intervals during the grazing period the length of 120 ungrazed leaves within the enclosure areas of each plot was measured, together with the length of 120 leaves recorded along three transects within each plot. Utilization was calculated for each plot as the difference in the mean length of ungrazed and grazed leaves, expressed as a percentage of the ungrazed length. Cattle were removed from the SNRG when the utilisation of the current season's growth of *M. caerulea* had reached an average of 50%. The data collection periods reported here ran from 3 June to 17 August in 2005, and 12 June to 1 September in 2006.

The cows and calves were allocated to treatments and plots on the basis of uniformity of liveweight and body condition score, with calves balanced for gender. All cattle were weighed at three-week intervals. Data were analysed using residual maximum likelihood (REML) with fixed effects of grazing treatment, time and their interaction (i.e. liveweight gain), and a random effect of individual animal number.

## Results and discussion

In 2005 the cows on all three treatments gained weight over the course of the measurement period (Table 1), but the Limousin cows grazing the SNRG swards gained significantly less than Limousins grazing the improved pasture. The following year the Limousin cows on the indigenous vegetation lost weight, whereas the Belted Galloways grazing the same sward type had the highest weight gain.

Table 1. Effect of grazing management and breed type on cow performance.

		Lim PP	Lim SNRG	BG SNRG	s.e.d.	Significance
2005	Initial weight (kg)	592	583	508	25.1	$P < 0.01$
	Weight change (kg/d)	0.46	0.13	0.24	0.089	$P < 0.01$
2006	Initial weight (kg)	623	618	480	22.1	$P < 0.001$
	Weight change (kg/d)	0.16	-0.27	0.41	0.151	$P < 0.001$

A statistically significant difference in the initial liveweight of the calves from the two breed types was evident in both years (Table 2), and the larger Limousin cross calves gained

significantly more total weight per day than the smaller Belted Galloway calves in both seasons. However, when initial metabolic liveweight was taken into account the pattern was different. In 2005 the growth rate per unit metabolic weight was significantly lower for the Limousin cross calves than the Belted Galloways calves when grazing the SNRG, with the growth rates of the Belted Galloway calves similar to that of the Limousin calves grazing the improved pasture. In contrast, during the 2006 season the growth rates of the Limousin and Belted Galloway calves on the SNRG swards were similar, and lower than that of the Limousin calves grazing the improved pasture.

Table 2. Effect of grazing management and breed type on calf performance.

		Lim PP	Lim SNRG	BG SNRG	s.e.d.	Significance
2005	Initial weight (kg)	110	110	57	7.9	$P < 0.001$
	Liveweight gain (kg/d)	1.33	1.26	0.83	0.030	$P < 0.001$
	Liveweight gain (g/kg initial metabolic LWT <sup>0.75</sup> )	41	37	41	1.5	$P < 0.01$
2006	Initial weight (kg)	117	116	69	15.0	$P < 0.001$
	Liveweight gain (kg/d)	1.35	1.17	0.84	0.032	$P < 0.001$
	Liveweight gain (g/kg initial metabolic LWT <sup>0.75</sup> )	40	36	35	2.1	$P < 0.05$

Calving in the spring is often recommended in marginal areas in order to synchronize maximum nutrient requirements in lactating cows with the period of higher herbage availability and quality. In these conditions, spring calving can reduce feeding costs and increase bioeconomic efficiency. The current experiment has indicated that good levels of performance can be achieved for suckler cows and calves on both improved ryegrass/white clover swards and *Molinia*-dominant SNRG. Although the liveweight gain of the Limousin calves on the SNRG was significantly lower than that of those on the improved pasture, they gained around 1.2 kg d<sup>-1</sup>. Likewise, although in 2005 the growth rate of the calves from the traditional breed was significantly higher than that of calves from the modern breed when the difference in initial live weight was taken into account, the 2006 results demonstrate that breeds such as the Limousin can perform as well as a traditional breed when grazing *Molinia*-dominant SNRG during early summer. It would appear that the higher liveweight gains of the calves on the Lim SNRG treatment in 2006 compared to 2005 was associated with greater mobilisation of body stores by the Limousin cows on that pasture.

## Conclusions

The results obtained to date indicate that both traditional and modern breeds of cattle can potentially be used to manage indigenous grassland communities such as *Molinia*-dominant SNRG for environmental gain e.g. without unduly compromising agricultural productivity.

## Acknowledgements

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# Bird communities on Swedish wet meadows – importance of management regimes and landscape composition

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## Abstract

Wet meadows in Sweden currently depend on management by grazing to maintain habitats for various breeding and migrating birds. The general aim of this paper is to analyse effects of different management regimes (grazing and late mowing), landscape composition and flooding on inland wet meadow bird fauna and flora. Five wet meadow areas in southern Sweden were used for a four-year survey of breeding birds and environmental variables. Analyses were performed both by ordination (bird community-level) and by nonparametric correlation (single species-level). Habitat openness was the most important factor for bird community composition, suggesting that clearing of woodlands situated close to meadows could be important. The bird community was also strongly related to the amount of flooding. In addition, measures for restoring water table variation also need to be considered. Effects of management were weaker, but several species showed positive associations with swards of intermediate height (5-30 cm), which are typical of mowing with late-season grazing.

Keywords: wet meadow, mowing, grazing, bird conservation

## Introduction

Management of wet semi-natural grasslands (i.e. wet meadows) by grazing or mowing is of great importance for the conservation of bird diversity. However, the area of wet meadow habitats has been greatly reduced because of drainage of wetlands and cultivation. Furthermore, habitat quality for breeding birds has declined due to earlier hay cutting and higher stocking of cattle. During the period 1990-2000, 39% of European bird species of inland wetlands declined in numbers (BirdLife International, 2004). Many Swedish wet meadow birds have declined in numbers during recent decades, and cessation of meadow management has been suggested to be the main cause. Wet meadow management currently depends on grazing animals, whereas mowing may be an important alternative management strategy in the future. In central Sweden species dependent on short swards (e.g. waders) and the Corncrake (*Crex crex*) (a species preferring tall swards) occur in the same meadow sites. In large meadow areas, it should be possible to combine management strategies that are beneficial for species preferring different management regimes. However, the relative importance of management, landscape structure and amount of flooding and other habitat characteristics for the meadow bird fauna is poorly known.

## Materials and methods

During 2001-2004 investigations of breeding wet meadow birds and habitat composition were performed yearly at five inland wetland areas in southern Sweden (Table 1). The meadow sites consisted of semi-natural vegetation (i.e. dominance of sedge (*Carex*) and wild grass (*Poaceae*) species). Within these areas a total of 137 wet meadow sites were censused. The meadows were managed according to the criteria of a conservation programme, i.e. mowing was usually done in the second half of July. The grazing season started between May 1 and June 5, with large variation between years and farmers. Management by grazing occurred on



101 sites, by mowing on 43 sites, by mowing with late season grazing on 29 sites, while 18 sites were unmanaged. Breeding meadow birds and habitat variables were surveyed each year according to a point census method (Hellström and Berg, 2001). A minimum distance of 400 m between census points was kept to reduce the risk of double counting. At each census point, management, habitat (e.g. flooding, sward height) and landscape characteristics (e.g. distance to different habitats) were mapped during bird censuses, but also collected from public statistics and compiled by use of GIS tools (see Gustafson, 2006). Ordination by Redundancy analyses (RDA) and Monte Carlo permutation tests (MCPT, with forward selection) were used (Biometris 1997-2003) in order to interpret general responses of the species community to management, habitat and landscape variables (environmental variables,  $n = 47$ ). Relations between abundance (breeding pairs) of single species and management and sward variables were investigated by nonparametric, partial Spearman correlation (mean values 2001-2004). Partial correlation was used to remove the effect of the main environmental gradients (variables correlated with ordination axes 1 and 2, see below).

Table 1. Description of the five meadow areas.

	Wetland area				
	Vattenriket	Roxen	Hjälstaviken	Fyrisån	Svartån
Location N/E	55°59' / 14°12'	58°29' / 15°33'	59°40' / 17°23'	59°50' / 17°39'	59°56' / 16°17'
Open meadow habitat (ha)	1400	200	210	50	670
No. of census sites	38	20	17	13	49
Water level variation (m)	1.4	1	1	0.5	0.7
Precipitation (mm)	6-700	6-700	5-600	5-600	7-800
Vegetation period (days with mean temp. > +5°C)	200	190	180	180	170

## Results and discussion

Habitat openness (abundance of trees and shrubs) was suggested by the RDA (Figure 1, axis 1 interset correlation = 0.49) and the MCPT to be the most important factor (F-ratio = 5.79,  $P < 0.01$ ) for composition of bird species ( $n = 33$ ). The second major factor that affected the bird community was wetness (amount of flooding in May 21-June 5), representing a gradient from dry to flooded meadows adjacent to shallow waters (RDA, axis 2 interset correlation = -0.48, MCPT, F-ratio = 5.41,  $P < 0.01$ ). The partial Spearman correlation analysis suggested 27 species to be significantly associated to management or sward structure variables. Although some passerine species (e.g. Thrush Nightingale *Luscinia luscinia*) seemed to prefer shrub or partly forested habitats, species preferring open meadows (e.g. Yellow Wagtail *Motacilla flava flava*) decreased strongly in total abundance even with a small amount of forest (0-5%) at the 250 m scale. Such small amounts of forest were common at the meadow areas (varying between 30% and 86% of the sites among areas). However, the single species analysis indicated that mowing in combination with late season grazing resulting in swards of intermediate height (5-15 cm), was preferred by many bird species. The avoidance of forested areas by several species dependent on short swards (continuous management), suggests that the most intensive management regimes (grazing and mowing combined with grazing) should preferably be targeted on landscapes with large open meadow areas without woodland, or where woodlands may be cleared. In contrast, less intensively managed meadows could be situated in partly forested areas since meadow birds associated with tall or periodically unmanaged swards did not seem to avoid areas adjacent to forests. A larger effort should be put into creating spatial and temporal variation in humidity of meadow sites, in particular to slow down the drying up after spring flooding.

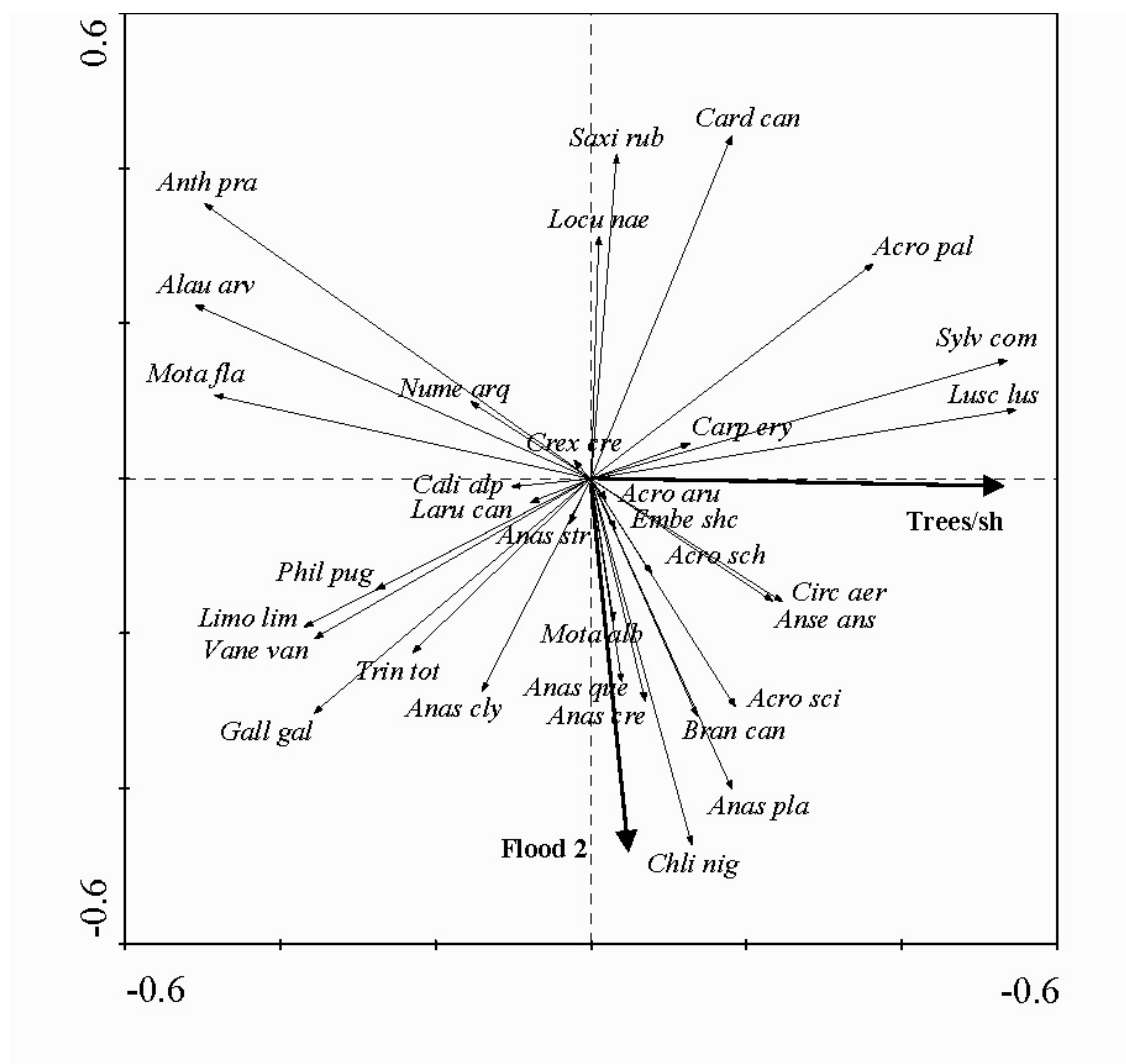


Figure 1. Redundancy analysis (RDA) of the bird community (abbrev. of scientific names: first four letters for genus and first three letters for species) with standardized species correlations. Length of a species' arrow indicates measure of fit ( $R$ ) with the ordination diagram (axis position: 1 = horizontal, 2 = vertical). Only environmental variables associated with significant axes are showed (Trees/sh = abundance of trees and shrubs, Flood 2 = amount of flooding in May 21-June 5).

## Acknowledgements

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# Stability of the greenery in different light conditions in the old park at Falenty, Poland

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## Abstract

This paper reports on an investigation of the effects of different light conditions on the stability, expressed by the underground to above-ground biomass ratio, of two grass mixtures sown in 2001 in the old park of the Palace-Park Complex at Falenty, Poland. The study was made in 2006 in two areas with different light conditions. Two grass mixtures (Dutch and Polish mixtures) were compared using two renovation methods (oversowing and full cultivation). All the examined grassy areas were characterized by higher underground than above-ground biomass, which provided evidence for the stability of these communities. Light conditions greatly affected plant biomass. The underground biomass was greater in the area with more intensive insolation.

Keywords: stability, biomass, insolation, country parks

## Introduction

Many precious country parks in historic palace-park complexes in Poland have been destroyed. These lands need appropriate protection, renovation and cultivation. Selection of appropriate mixtures to renovate them is very important. Well-kept greenery emphasizes the beauty of other plants and of historic buildings. Nevertheless, it is difficult to achieve good plant status in a park because of diverse light conditions (Starc *et al.*, 1995; Mahallati, 1998). Preservation of plant structure with underground biomass greater than the above-ground biomass favours the stability and durability of plant communities under Polish climatic conditions (Stańko-Bródkowa, 1989). The aim of this study was to investigate the relationship between the underground and above-ground biomass, and the distribution of underground biomass in particular layers of renovated greenery located in areas with different light conditions in Falenty Park.

## Material and methods

The study on plant stability was carried out in 2006 in the 17th-century Palace-Park Complex at Falenty near Warsaw, Poland. An experiment with renovation of abandoned park vegetation was set up in autumn 2001 and included two grassy areas (area 1 and area 2) located in different light conditions. Area 1 receiving higher insolation (mean  $506 \text{ W}\cdot\text{m}^{-2}$ ) and was located in a place overgrown by *Tilia cordata* Miller. Area 2 was characterized by less intensive sunshine (mean  $278 \text{ W}\cdot\text{m}^{-2}$ ) and the dominating trees were *Quercus robur* L. Light conditions were defined based on total radiation measured systematically during the whole vegetation period with a thermostat on sunny days and with albedometer on cloudy days. Two grass mixtures, a Dutch mixture produced by the Barenbrug company (M1) and a Polish mixture (M2) established using two renovation methods (oversowing with a verticulator and full cultivation with a rototiller) were used. M1 contained *Lolium perenne* L. (Figaro 16%, Stadion 14%), *Poa pratensis* L. (Nimbus 15%) and *Festuca rubra* L. (Bargena 30%, Barskol 10%, Bernica 15%), and M2 contained *Lolium perenne* L. (Nadmorski 50%), *Poa pratensis* L. (Balin 10%), *Festuca rubra* L. (Areta 10%, Leo 20%) and *Festuca ovina* L. (Spartan 10%). The treatments had 4 replications and each plot size was  $4 \text{ m}^2$ .

The above-ground biomass in g dry matter (DM) per 1 m<sup>2</sup> was estimated as a sum of mean yield in 2006 and stubble mass. The underground biomass in g DM per 1 m<sup>2</sup> was measured in the root layer (0-20 cm) separated into four layers (0-5, 5-10, 10-15 and 15-20 cm). The underground biomass was taken with a special cylindrical instrument ( $\phi$  8 cm, height 17 cm). Samples of underground biomass were rinsed on sieves of a mesh size 0.80 and 0.43 mm and manually cleaned later. The floristic composition was determined by botanical analyses in 2004. The data were processed statistically using multiway classification analysis of variance. Differences among treatments were tested with LSD test.

## Results and discussion

The structure of plant biomass was analysed in 2006, the fifth year after sowing both mixtures. The occurrence of strongly developed root systems in each studied variant was confirmed (Figure 1). The underground biomass in the less insolated area was significantly greater (1.5-2.3 times) than in the area with higher insolation. The largest underground biomass was produced by the Polish mixture in the variant with oversowing (1764 g m<sup>-2</sup>), and by the Dutch mixture after full cultivation (1629 g m<sup>-2</sup>). The smallest underground biomass was found in the area with higher insolation in the oversowing variant for both mixtures (658 g m<sup>-2</sup> – M1, and 774 g m<sup>-2</sup> – M2). The distribution of underground biomass in particular layers did not differ from the typical structure for grassy areas, and the main biomass of underground organs was found in the sward layer (0-5 cm). The sward layer in the area 2, with less intensive sunshine, contained 1.3-3.0 times more biomass than that in area 1, for the same variants of renovation methods and seeds mixtures. It contained 48-59% of total underground biomass in area 1 and 43-64% in area 2. The percentage division of biomass among particular layers in both areas was similar. For comparison, the sward layer in permanent grasslands at Jaktorow contained over 60% of total underground biomass (Rutkowska *et al.*, 1980). In both areas there were no significant differences in the amounts of underground biomass between mixtures for particular renovation methods (Table 1).

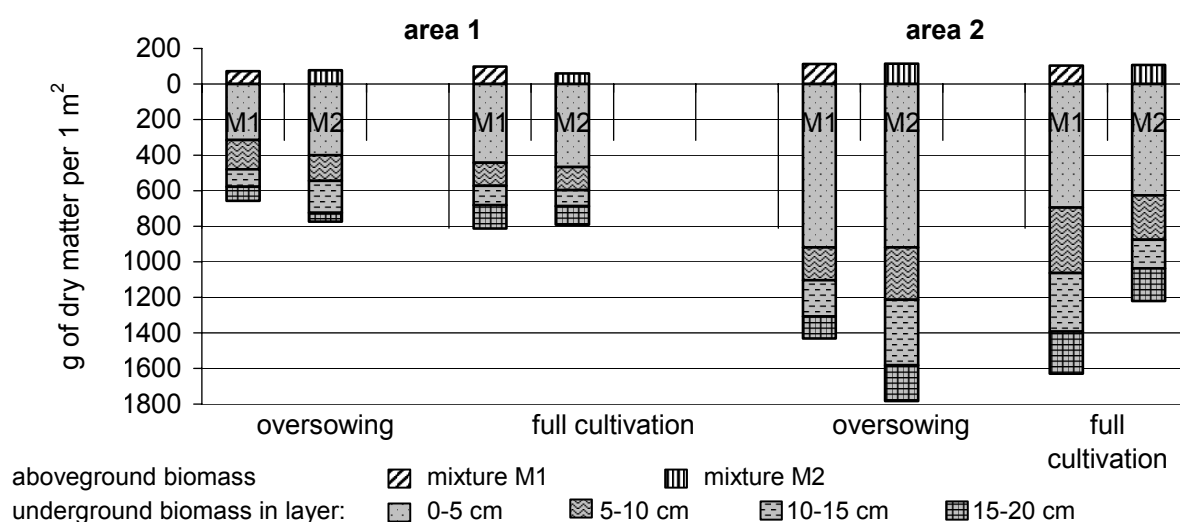


Figure 1. Biomass (g DM m<sup>-2</sup>) in relation to seeds mixture (M1, M2) and establishment method for areas with high (1) and low (2) insolation in the park experiment.

The ratio of underground to above-ground biomass which occurred in all variants was characteristic of permanent communities (Figure 1). The underground biomass was

considerably larger (8-16 times) than above-ground biomass. Higher ratios were found in the less insolated area. The highest ratio was noted for the Polish mixture in places renovated by oversowing and for the Dutch mixture applied in full cultivation. The lowest ratio was found in the more insolated area for the Dutch mixture at both renovation methods. The above-ground biomass was very small because of the characteristics of the lawn varieties sown in mixtures and adverse weather conditions (long rainless periods with high temperatures) in summer 2006. The largest above-ground biomasses were found in less insolated area. The above-ground biomasses of two mixtures in places renovated by oversowing and by full cultivation were similar. The larger underground biomass found in area 2 resulted in the increase of the underground to aboveground biomass ratio. Analysis of the floristic composition in park areas showed that grasses predominated in above-ground biomass of the area with more intensive solar radiation, while dicotyledons dominated in less insolated areas. A substantial share (mean 83%) of low grasses, mainly *Lolium perenne*, in the park sward, and the smallest number of plant species were recorded in places renovated by full cultivation. Low grasses contributed 70-80% in oversown places.

Table 1. MANOVA and least significant differences for effect of light conditions and plant mixture and location of layer on underground biomass ( $\text{g m}^{-2}$ ) in the park experiment.

Variation	df	Oversowing with verticulator			Full cultivation with rototiller		
		MS	F <sub>emp</sub>	LSD <sub>0.05</sub>	MS	F <sub>emp</sub>	LSD <sub>0.05</sub>
light conditions (Lc)	1	794995.14	30.29**	82	388440.56	14.24**	84
mixture (M)	1	54697.52	2.08	ns	46332.56	1.70	ns
layers (La)	3	887779.14	33.82**	154	564975.46	20.72**	157
interaction Lc/M	1	13835.64	0.53		37636.00	1.38	
interaction Lc/La	3	205296.43	7.82**		9687.69	0.36	
interaction M/La	3	7238.47	0.28		3623.19	0.13	
interaction Lc/M/La	3	9856.68	0.38		2791.46	0.10	
Error	45	26249.12			27271.86		

\*\* $P < 0.01$ ; ns – not significant.

## Conclusion

The study on the structure of plant biomass provided confirmation that all the examined sown swards had the features of a permanent community with some dynamic equilibrium stability. There was a predominance of underground biomass over above-ground biomass. The weight of underground organs in the sward layer comprised 43-64% of the total below-ground biomass. Generally, in 2006 the amounts of above-ground biomass were small because of the characteristics of the lawn varieties of plant sown in the mixtures and the adverse weather conditions in the summer of 2006. The location of the examined grassy areas, in terms of their different light conditions, affected the stability of the park greenery.

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# Foraging behaviour and diet composition in cattle on semi-natural grasslands

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## Abstract

Optimal grazing management is important to maintain biodiversity in semi-natural grasslands. The aim of this study was to evaluate the effects of breed, season and pasture moisture gradient on foraging behaviour, faecal plant fragment composition and defoliation of competitive plant species by cattle on semi-natural grasslands. Heifers of the Swedish traditional breed Väneko and the commercial breed Charolais grazing semi-natural grasslands during spring, summer and autumn of 2004 were studied. The Väneko heifers had a higher activity than the Charolais heifers ( $P = 0.006$ ). As the grazing period progressed, time spent grazing ( $P < 0.05$ ) and efficiency of grazing ( $P < 0.001$ ) increased. Proportions of Cyperaceae (sedges and rushes) and herbs were highest in the spring ( $P < 0.001$ ), whereas the proportion of *Festuca rubra* (red fescue) was highest in the autumn ( $P < 0.01$ ). Defoliation intensity of *Filipendula ulmaria* (meadowsweet) and *Alnus glutinosa* (alder) increased as the grazing period progressed ( $P < 0.05$ ). On average over seasons, 28% of the herbage mass was found in the wet areas, dominated by *Deschampsia cespitosa* (tufted hairgrass), whereas only 8% of the grazing occurred there. In conclusion, foraging behaviour and diet composition were affected more by season than by breed, and wet areas were avoided for foraging.

Keywords: breed, season, moisture gradient, grazing, defoliation, nature conservation

## Introduction

Optimizing grazing management is important to maintain biodiversity in semi-natural grasslands. Behavioural theories predict that: 1) livestock of traditional breeds will forage less selectively and more actively than commercial breeds, 2) animals will forage more effectively in the late than the early grazing period, and 3) animals will avoid foraging in areas with low-nutrient herbage. The objectives of this study were to determine the effects of breed, season and pasture moisture gradient on the extent and location of foraging, activity, faecal plant-fragment composition, and defoliation of competitive plant species by cattle kept on heterogeneous semi-natural grasslands.

## Materials and methods

The experiment was conducted at The Götala Research Station in south-western Sweden from May to October 2004 on 18 hectares of heterogeneous, mainly open, semi-natural grasslands. Two groups of heifers, 12 of the traditional Swedish breed Väneko and 12 of the commercial Continental breed Charolais, were allocated into three enclosures per breed. Each enclosure consisted of 20% dry, 60% mesic and 20% wet areas. Dominating plant species were *Festuca ovina* (sheep's fescue), *Deschampsia flexuosa* (wavy hairgrass), *Nardus stricta* (matgrass) and several herb species in dry areas. *Festuca rubra* (red fescue) and *Deschampsia cespitosa* (tufted hairgrass) and herbs dominated in mesic areas. *D. cespitosa* and Cyperaceae (sedges and rushes) were prominent in wet areas.

In spring, summer and autumn, the behaviour of every heifer was recorded for 24 h using automatic behaviour recorders (Rutter *et al.*, 1997; Rutter, 2000), and animal positioning and activity were recorded using GPS receivers and accelerometers (GPS Plus 2, Vectronic Aerospace GmbH, Berlin, Germany). At the same occasions, faeces samples were collected for microhistological examinations of plant fragments (Garcia-Gonzalez, 1984). Furthermore, defoliation of the competitive plant species *Filipendula ulmaria* (meadowsweet), *Juncus effusus* (soft-rush), *Alnus glutinosa* (alder) and *D. cespitosa* was recorded in plots in the wet areas. Pasture herbage was analysed for contents of crude protein (CP), neutral detergent fibre (NDF) and metabolizable energy (ME). At the end of the grazing period, the pasture was visually examined to assess if the sward heights were short enough to ensure that no litter had accumulated on the sward.

Statistical analysis was performed by using the procedure Mixed with repeated measurements for data on behaviour, activity, plant fragments and defoliation (SAS, 2001). For faecal plant fragments, Bonferroni tests were used to correct for false significant effects (Hochberg, 1988). The procedure t-test was used for foraging areas and the procedure Genmod for visited plots (SAS, 2001).

## Results and discussion

The Väneko heifers had a higher activity than the Charolais heifers ( $P = 0.006$ ), but no main effects of breeds on location of grazing or faecal plant-fragment composition were found. However, Väneko heifers defoliated more *F. ulmaria* in the spring ( $P = 0.009$ ) and they increased their defoliation of *A. glutinosa* more during the grazing period ( $P < 0.001$ ) than Charolais heifers. A higher activity in cattle of traditional compared to commercial breeds is consistent with previous findings (Sæther *et al.*, 2006).

Season affected both behaviour and diet in the heifers. The heifers spent more time grazing in autumn (42.5% of the day) than in spring (38.5%;  $P = 0.006$ ) and summer (38.9%;  $P = 0.014$ ) and the efficiency of grazing (i.e. proportion of eating during grazing bouts) increased over the grazing period ( $P < 0.001$ ). The prolonged grazing time in autumn can be explained by decreased concentrations of CP and ME in the herbage in the late grazing period ( $P < 0.001$ ) but do not explain the increased efficiency in grazing. Anti-predator theory predicts that large herbivores avoid foraging in darkness due to a perceived risk of predation (Rutter, 2006). Fewer daylight hours in the autumn may explain why the grazing was more effective in autumn than earlier in the grazing period, i.e. the extent of daylight hours limited the time available for grazing. Shorter distances travelled and lower activity of the animals in the autumn ( $P < 0.05$ ) also support the anti-predator theory.

Proportions of Cyperaceae and herbs in faecal plant fragments were higher in the spring than later in the grazing period ( $P < 0.001$ ), whereas the proportion of *F. rubra* was highest in the autumn ( $P < 0.01$ ). Defoliation intensity of *A. glutinosa* and *F. ulmaria* increased as the grazing period progressed ( $P < 0.05$ ). More woody plants in the diet in late season is supposed to be due to a lower nutrient quality in grasses and herbs during autumn than earlier, whereas nutrient concentrations in leaves are relatively constant over the grazing period (Ganskopp *et al.*, 1999). Consequently, grazing livestock are better at controlling pernicious brushwood in autumn than in spring.

On average over seasons, 28% of the herbage mass was found in the wet areas, whereas only 8% of the grazing occurred there. Furthermore, the proportion of eating during grazing bouts was lower (73.9%;  $P < 0.001$ ) in wet areas than in dry (80.1%) and mesic (79.6%) areas. The avoidance of wet areas resulted in only 4% of faecal plant fragments being composed of Cyperaceae and defoliation of only half of the *D. cespitosa*- and *J. effusus*-plots. Consequently, at the end of the grazing period, the grazing pressure was assessed as having been weak in the wet areas, whereas it was satisfactory in the dry and mesic areas. Herbage in the

wet areas had lower concentration of CP ( $P = 0.036$ ) and higher concentration of NDF ( $P = 0.011$ ) than herbage in dry areas. Cattle actively forage for nutritious feed (Launchbaugh and Howery, 2005) and their avoidance of foraging in wet *D. cespitosa*- and *Cyperaceae*-dominated semi-natural grasslands may constitute a challenge when managing these areas (Krahulec *et al.*, 2001). Dividing heterogeneous grasslands with fences to direct robust animals to these areas in early spring, thereby aiming to delay the maturation with accompanied decreased digestibility of these plants, may be necessary to obtain satisfactory management of these areas.

## Conclusions

The extent and location of foraging by cattle on semi-natural grasslands, as well as their diet composition, were more affected by season than by breed. The cattle browsed more leaves from pernicious woody species such as *A. glutinosa* in autumn than earlier in the grazing period. Wet *D. cespitosa*-dominated areas were avoided for foraging throughout the grazing period. Strategic use of a limited number of grazers might be to direct them to *D. cespitosa*-dominated areas in spring, whereas control of brushwood can be postponed to autumn.

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# Effects of social environment on grazing behaviour and liveweight changes in first-season-grazing dairy calves

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## Abstract

The aim of this study was to investigate if the foraging behaviour of naïve calves turned out to pasture was more effective, and losses in liveweight thereby diminished, when the calves were accompanied by grazing-experienced cattle. Twenty-six first-season-grazing dairy calves were turned out to semi-natural grasslands allocated into groups with or without the company of second-season-grazing steers. The behaviour of the calves was automatically recorded for 24 h during their first day on pasture, and after having been on pasture for one month, and the average liveweight changes were calculated for their first month at pasture. Calves turned out to pasture in the company of experienced conspecifics were more active during grazing as measured by accelerometer sensors (score 132 vs. 117,  $P = 0.023$ ) than calves turned out to pasture in groups of their own. However, the higher grazing activity was not confirmed by longer grazing time or diminished liveweight losses. In conclusion, the company of grazing-experienced conspecifics resulted in higher grazing activity in naïve calves, but no effects on liveweight change were found.

Keywords: naïve grazer, turn-out, foraging, activity, weight gain, social learning

## Introduction

Social learning plays an important role in the development of foraging skills of domestic ruminants (Launchbaugh and Howery, 2005). The dam is the best social model for the young calf, but in gregarious species other dominant conspecifics are also of significance for learning diet selection and for developing feelings of security (Green, 1992; Bailey *et al.*, 2005). Dairy calves in their first season of grazing are turned out to pasture without the company of their dams. Anxious and inexperienced at foraging in this new environment, these naïve grazers might have an inefficient foraging behaviour, resulting in suboptimal pasture management and liveweight gains (Ksiksi and Laca, 2000). The aim of this study was to evaluate the effects of turning out first-season-grazing dairy calves together with experienced conspecifics on the calves' behaviour and liveweight changes during their first month at pasture.

## Materials and methods

The experiment was conducted at The Götala Research Station in south-western Sweden from May to June 2005 on 18 hectares of heterogeneous, mainly open, semi-natural grasslands dominated by tufted hairgrass (*Deschampsia cespitosa*). Twenty-six steer calves of the dairy breed Swedish Red with an initial average liveweight of 260 kg (SD 37) were studied. The calves were allocated into six enclosures on the pasture, three to six calves in each enclosure depending on enclosure area. In three of the enclosures, the calves were accompanied by two

second-season-grazing steers with experience from similar pasturelands, whereas the calves in the remaining three enclosures were grazing in groups of their own.

On three calves per enclosure, jaw movements were recorded automatically using IGER Behaviour Recorders (Rutter *et al.*, 1997) during the first 24 h on the pasture and during 24 h after having been on pasture for a month. Jaw movements were defined as eating or ruminating jaw movements using the Graze software (Rutter, 2000). Eating time was defined as the time spent performing eating jaw movements, and grazing time was defined as eating time plus pauses between eating bouts shorter than 7 min (Rutter, 2000). The proportion of grazing time that consisted of eating bouts was calculated and defined as grazing efficiency. Ruminating time was defined as time performing ruminating jaw movements plus pauses between ruminating jaw movements shorter than 20 s. Animal position fixes were recorded every 15 s using collars equipped with GPS receivers (GPS Plus 2, Vectronic Aerospace GmbH, Berlin, Germany). Data for activity measurements was recorded by two orthogonally orientated accelerometer sensors on the collars and scaled to numerical values from 0 (no activity) to 361 (maximum activity). Average activity scores were calculated for 24 h and separately for time periods animals spent grazing, ruminating and idling. Animal data were merged with pasture area data using the GIS software ArcMap (ESRI, 2006) and travelling distances of each animal were calculated. Liveweight changes of all calves were calculated for the first month on pasture.

Statistical analyses were undertaken for grazing, activity, distance travelled and liveweight changes using the procedure Mixed (SAS, 2003). Data were averaged over animals for each enclosure resulting in three replicates per treatment. The statistical model included two fixed factors: social environment (company of older steers or not), and time (at turn-out and after one month) with enclosure as random factor.

## Results

Averaged over the two times, calves in company with grazing-experienced steers had a higher activity during grazing than calves kept in groups of their own (Table 1). No effects of social environment on liveweight change during the first month on pasture could be found (average of 0.30 kg day<sup>-1</sup>). Nor could any interactions between social environment and time be found.

Table 1. Behaviour of groups of first-season-grazing calves ( $n = 3$ ) on semi-natural grasslands with or without company of grazing-experienced steers at turn-out to pasture and after one month on pasture; SEM is standard error of the mean.

	Company		Time		SEM	$P^a$	
	With	With-out	Turn-out	One month		Com-pany	Time
Grazing (% of day)	44.4	41.2	44.8	40.8	2.4	ns	ns
Ruminating (% of day)	25.0	21.8	18.8	28.0	1.6	ns	0.003
Idling (% of day)	30.7	37.0	36.5	31.3	3.3	ns	ns
Grazing efficiency (%) <sup>b</sup>	72.1	68.7	63.4	77.4	2.6	ns	0.005
Total activity (score)	86	80	95	70	7	ns	0.039
Grazing activity (score)	132	117	111	137	3	0.023	0.002
Ruminating activity (score)	24	25	29	21	2	ns	0.042
Idling activity (score)	72	70	107	35	17	ns	0.015
Distance travelled (km)	15.3	14.0	17.6	11.7	1.1	ns	0.007

<sup>a</sup>Level of significance for effect of company or time: ns = non-significant,  $P > 0.10$ .

<sup>b</sup>Proportion eating bouts during grazing time.

Calves were more active, travelled longer distances and ruminated less during their first day on pasture than after having grazed for a month (Table 1). However, activity during grazing, and grazing efficiency were lower during the first day on pasture than after having grazed for a month (Table 1).

## Discussion

Calves kept together with grazing-experienced steers as social models were more active during foraging than calves kept without this company, whereas grazing time, grazing efficiency and liveweight change were similar between the two treatments. Unrelated experienced conspecifics as social models for inexperienced ruminants during foraging learning has been experimentally investigated with inconsistent results. In a study of Ksiksi and Laca (2000), calves in company with grazing-experienced steers found more feed than calves without this company, whereas Bailey *et al.* (2000) failed to improve foraging ability in heifers by using experienced leaders. Thorhallsdottir *et al.* (1987) found that lambs reared with social models consumed similar amounts of feed within a shorter time period than orphan lambs. The absence of obvious differences between the two treatments in the present study are in agreement with Ganskopp and Cruz (1999) and Ksiksi and Laca (2000), who found that calves' essential selection of herbage takes place within a few minutes on pasture, with the result that, within a few days, naïve grazers without social models are able to search for feed better than would be expected by chance.

## Conclusions

Calves turned out to pasture accompanied by experienced conspecifics were more active, as measured by accelerometer sensors during grazing, than calves grazing in groups of their own. However, the increased activity was not confirmed by increased grazing time, grazing efficiency or liveweight gains.

## Acknowledgements

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# The effect of grassland renovation and break up on nitrogen losses

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## Abstract

This paper focuses on a series of experiments we conducted over an 8-year period (1999 till now) on sandy soils concerning the renovation of grassland and its break up for arable use. Our main interest was the effect of such break up or renewal on the environment, namely nitrate leaching, soil mineral nitrogen (SMN) and related soil processes. Among the factors we considered were: the influence of season of renewal, of previous N management (including clover), of N fertilization for the new sward, and of technique of renovation; the influence of the crop and N management following a grassland break up, and of soil characteristics on SMN and nitrate leaching. We also compared permanent grassland with long intervals of renewal, or break up with regular forage farming using Italian ryegrass and maize. Our findings suggest that spring renovation is preferable to autumn renewal, and actual N application should be reduced in order to minimize nitrate losses. After break up for arable use, spring barley followed by a catch crop was favourable to the cultivation of silage maize in storing mineralized N and controlling N leaching.

Keywords: grassland, renovation, break up, nitrogen, leaching

## Introduction

Grassland renovation and grass-arable rotations or break up for permanent arable use are of major concern for the economic benefit of dairy and forage farms and for their impact on the environment (Conijn and Taube, 2007). Renewal is mainly due to decline in yield and nutritive value or to sward damage. The temporal or permanent conversion to arable land is often motivated by a shift to ley farming, socio-structural reasons and effects of the CAP reform. Changes in land management affect the environment on different scales. With the focus on management options in water protection areas, we were mainly interested in the effect of sward destruction on nitrogen leaching at the field scale.

## Materials and methods

We present results from several experiments in northwest Germany on sandy soils which include deep-ploughed soils from podzols and a plaggen soil. Among the factors considered were the previous levels of N fertilization, the new N level, choice of the following crop, and season of renewal or break up. Renewal was carried out with a permanent seed mixture based mainly on *Lolium perenne*, with or without white clover. Arable crops were silage maize and spring barley with a catch crop and a ley mixture based on *Lolium multiflorum*, with or without red clover. Renewed grasslands were previously mown and invariably used as cut grasslands again. In all experiments we used suction cups to sample for nitrate concentrations and to subsequently model N leaching losses; we also did regular soil sampling to determine soil mineral N from a depth of 0-30 cm to 0-90 cm.

## Results and discussion

Although N leaching losses and soil mineral N in autumn were influenced by different weather conditions during the experimental years, especially rainfall during winter, our results indicate some clear trends (Table 1).

Table 1. Soil mineral N (SMN) and N leaching losses (suction cups) for different scenarios of grassland renewal and break up from a series of experiments (1994-2006) in northwest Germany on predominantly sandy soils; values are averages derived from different experiments. SMN and N leached are for the first autumn/winter following renewal or break up, values in brackets are from the second crop and/or second autumn/winter.

up, values in brackets are from the second crop and/or second autumn/ winter.					
Crop following renewal or break up	Previous management and mode of cultivation	Previous N level [N kg/ha]	New N level [N kg/ha]	SMN autumn [N kg/ha]	Leached N [NO <sub>3</sub> -N kg ha]
Reference					
Permanent Grassland (PG)	9-year-old sward; 4 cuts for silage; data from 6 years	160	-	26	8
		320	-	45	17
PG + white clover		0	-	38	29
Late summer renewal / break up					
Grass	9-year-old sward; rotary tillage	0	0	58 (30)	35 (5)
		320	0	80 (21)	55 (3)
Grass permanent mixture	> 15-year-old grassland - 2 years arable: spring barley	120-160 / 30 for catch crop	0 (240)	65 (18)	49 (9)
Grass ley mixture	and maize; catch crop; ploughed	120-160 / 30 for catch crop	0 (360)	47 (23)	21 (34)
Spring renewal / break up					
Grass	9-year-old grassland; rotary tillage,	320	0	18 (19)	1 (2)
		320	160	24 (26)	2 (3)
		320	320	37 (67)	10 (45)
Spring barley + mustard – (maize)	> 15-year-old grassland; ploughed	120	0	26 (25)	45 (28)
		120	120 (160)	40 (182)	81 (177)
Maize – (maize)		120	0	113 (14)	118 (17)
		120	160 (160)	163 (90)	225 (101)

Generally, the effect of grassland renewal on N leaching losses is relatively small compared with the conversion of grassland into arable land and regular soil tilling. The results of our renovation experiments show that grassland renewal in late summer can lead to increased N leaching losses during the following winter, especially under intensive management and with N surpluses before the date of renewal. To minimize this risk, the date of renewal should be moved from late summer/autumn to spring. In this case, the level of previous N fertilization and technique of renewal are of less importance. If a renewal in late summer/autumn is inevitable because of soil conditions, special attention should be given to reduce N input before renewal. Relatively small N leaching losses soon after renovation and thereafter are also reported by Eriksen (2001) and Shepherd *et al.* (2001). After renovation of mown grassland there is no requirement to reduce nitrogen fertilization far below a level that can be taken up and utilised by the crop (Seidel *et al.*, 2007). N mineralization is highest in the first two years after grassland destruction, with the first year accounting for 70-90% of the cumulative mineralization (Vertès *et al.*, 2007).

When older grassland (> 15 years) from a mixed use of grazing and cutting was ploughed for arable use in spring, nitrate leaching was greatly increased. After grassland break up, N fertilization to the succeeding crops with amounts of N following common practice, resulted in almost doubling SMN and N leaching losses compared with the non-fertilized plots.

Eriksen (2001) also found that the current quantity of N fertilizer applied had a greater impact on SMN than the previous management of the grassland. In our experiments, the effects of the grassland break up on N dynamics were still strong during the second year. Results for the first two years indicated that the crop sequence spring barley+catch crop - maize compared to maize - maize was better suited to mitigating N leaching losses. The catch crop (yellow mustard) was essential and more than 50 kg N ha<sup>-1</sup> were taken up into the above-ground plant mass, disregarding stubble and roots. Mineral N application needs to be reduced to at least 60-100 kg N less than usual (Neuens and Reheul, 2002). From the second year onwards, a well adjusted N fertilization level seems to be more effective than the choice of the crop; silage maize, with its high potential for N uptake, is a possible crop. When older grassland is to be turned into permanent arable land, cultivation of Italian ryegrass for the first 1-2 years might be a good start. However, there can be some agronomic problems with spring renovation or break up: the first growth period is shortened, on wet sites tillage will be delayed, annual dicots compete with the new grasses, and pests like wire worms may infect the following crops.

## Conclusions

If a change from permanent grassland to arable land is necessary and soil and site conditions allow this, the time of break up, the choice of the crop and N fertilization should ensure that the N from the inevitable mineralization is utilized effectively and buffered. The negative effects of a grassland break up are hardly ever completely suppressed during the first year. This might be different in a grass-arable rotation where grassland is rarely older than 3 years or when grassland is renovated in spring with moderate N fertilization during the first year.

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# Influence of the site moisture and trophic conditions on the floristic diversity of meadow communities

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## Abstract

In this research we determined the influence of soil moisture and soil fertility on some elements of the floristic diversity of selected meadow communities. Reduced site moisture content was found to favour increased concentrations of available forms of nitrogen, and decreased magnesium and sodium in soils. Reduced moisture content and the content of the available forms of elements, contributes to gradual transformations in the floristic composition of communities and development of variants exhibiting lower species wealth in comparison with the typical variant. Communities of the *Phragmitetea* class were characterized by a greater number of plant species and a higher floristic diversity index, whereas those of the *Molinio-Arrhenatheretea* class by a smaller number of plant species and a low Shannon-Wiener index,  $H'$ .

Keywords: permanent grassland, floristic diversity, site conditions

## Introduction

Site conditions exert a significant impact on the development of meadow communities. This is particularly true with regard to the site moisture content and trophic conditions which initiate succession processes leading to transformations in the floristic composition of communities. It is known that some species of meadow plants respond to increased quantities of nitrogen released from the soil as a result of decreases in their moisture content. This process leads to reduction in diversity of the species composition of meadow communities or even to their uniformity. The effect of the concentration of available forms of potassium, magnesium or sodium on the floristic diversity of plant communities is less known. Many researchers emphasize that phosphorus and nitrogen are among the elements which influence most dramatically the floristic variability of many plant communities (Janssens *et al.*, 1997; Critchley *et al.*, 2002; Böhner, 2005). The aim of this research project was to determine the influence of site conditions, in particular soil moisture content and soil fertility, on some elements of the floristic diversity of selected meadow communities.

## Materials and methods

The effect of site moisture variations and changes in the total soil nitrogen and available forms of nitrogen and phosphorus, as well as potassium, magnesium and sodium, was assessed on the basis of the analysis of the floristic wealth (number of species in a phytosociological survey), floristic diversity (the value of the Shannon-Wiener index,  $H'$ ) of the identified meadow communities. The following parameters were determined in the course of the performed experiments: the soil moisture (F) and the content of total nitrogen (N) by Ellenberg's index; phosphorus and potassium, by the Egner-Riehm method in mineral soils (in the solution of calcium lactate) and in HCl in organic soils; magnesium and sodium by the Schachtschabel's method in mineral soils (in solution of calcium chloride) and in HCl in organic soils; ammonium and nitrate nitrogen (using a FIAstar<sup>TM</sup>5000 apparatus, FOOS Company, Sweden).

## Results and discussion

From among the abiotic site factors affecting the floristic diversity of meadow communities, moisture content appears to play a major role. Each change of this parameter leads to changes in soil trophic properties. Together with the decrease in soil moisture content, the content of total nitrogen in them also declines. Organic soils of marshy sites under the communities from the *Phragmitetea* class are rich in total nitrogen. Post-marshy or mineral soils on which communities from the *Molinio-Arrhenatheretea* class are found (in sires with variable or moderate moisture content) are characterized by a lower concentration of total nitrogen. However, in such conditions, a higher content of available forms of nitrogen (i.e. nitrate and ammonium forms) are determined and their presence is associated either with the mineralization of nitrogen bound in organic forms, or with the application of mineral fertilization (Table 1).

Table 1. Dependence of the floristic diversity of meadow communities of site conditions.

Syntaxa	Ellenberg's index		Content kg ha <sup>-1</sup>		Content mg 100g <sup>-1</sup> of soil			Number species in relevé	H'	
	F	N	N-NO <sub>3</sub>	N-NH <sub>4</sub>	P	K	Mg			
Phragmitetea – communities of marshy communities										
Phragmitetum australis	9.2	6.6	46.2	46.9	24.3	2.7	2.7	12.2	12	1.23
Phalaridetum arundinaceae	8.0	6.0	63.5	63.8	15.8	4.4	5.3	11.5	19.5	1.75
Molinio-Arrhenatheretea – seminatural and anthropogenic communities of non-marshy sites										
Com. Agrostis stolonifera - Potentilla anserina	7.2	5.5	119.1	143.9	3.9	5.9	9.2	27.1	19	1.69
Com. Deschampsia caespitosa	6.4	3.8	125.7	32.4	3.7	5.1	7.0	17.9	22	1.92
Lolio-Cynosuretum Arrhenatheretum elatioris	5.8	5.3	204.6	144.7	4.0	11.3	8.8	26.5	25	2.58
Com. Poa pratensis-Festuca rubra	5.2	4.9	108.8	91.0	6.9	7.7	7.2	15.4	21	2.71
	5.1	4.5	247.8	94.7	1.7	5.7	5.9	17.6	25	2.05

The content of other elements in the soils of the examined communities assumes certain regularity. Low potassium content in the soils of the examined communities deserves attention. Soils of the communities of the *Phragmitetea* class are characterized by very high phosphorus content and lower concentrations of magnesium and sodium. A reverse correlation was recorded in the case of communities of the *Molinio-Arrhenatheretea* class. Hence, together with the decrease in the site moisture content, the content of phosphorus increases, while that of magnesium and sodium declines.

Both soil moisture content and their trophic properties contribute to gradual transformations in the floristic composition, which affects the diversity of meadow communities. The consequence is the development of intermediate communities differing with regard to their floristic wealth in comparison with typical forms. The drying of marshy sites, the increase in the phosphorus and potassium content as well as the decrease in the concentrations of magnesium and sodium in the soils of the communities of the *Phragmitetea* class all lead to the improvement in the floristic diversity. Species of wider ecological scale, characteristic of sites of variable moisture content, begin to appear in patches of the existing communities (Table 2). A reverse process can be observed in the case of the communities from the *Molinio-Arrhenatheretea* class. Variants of associations developed in the conditions of lower moisture content are characterized by a lower floristic wealth associated with the dominance



of some plant species, primarily grasses, characterized by lower requirements connected with moisture, e.g. *Festuca rubra*, *Poa pratensis* (Table 2).

Table 2. Influence of the moisture content on the site trophic properties and floristic diversification of selected associations.

Reaction of selected associations.										
	Ellenberg's index		Content kg ha <sup>-1</sup>		Content of available forms mg 100g <sup>-1</sup> of soil				Number of plant species in relevé	H'
Variant	F	N	N-NO <sub>3</sub>	N-NH <sub>4</sub>	P	K	Mg	Na		
<i>Phalaridetum arundinaceae</i>										
Wet	8.4	6.4	47.7	59.0	9.1	9.7	68.9	14.4	8 (4-12)	1.2
Typical	6.9	6.1	96.2	92.5	8.6	5.4	89.8	13.5	12 (9-15)	1.5
Drying	6.4	5.9	115.6	124.7	10.0	10.1	47.6	9.3	21 (18-25)	2.3
<i>Arrhenatheretum elatioris</i>										
Humid	6.3	3.6	104.6	78.7	6.0	3.5	64.2	19.9	18 (11-38)	2.5
Typical	5.5	3.9	113.0	43.2	3.0	3.0	51.3	16.7	38 (23-49)	3.4
Drying	4.1	4.1	130.5	52.1	9.9	4.6	49.0	14.0	24 (20-34)	2.9
<i>Lolio-Cynosuretum</i>										
Humid	6.8	5.2	222.1	96.4	2.0	5.4	78.2	29.7	17(12-19)	1.9
Typical	5.7	5.0	302.4	124.1	3.2	4.1	52.8	21.9	26 (17-31)	2.8
Drying	4.9	4.2	211.1	219.1	4.0	5.7	37.5	14.3	23 (15-27)	2.2

## Conclusions

The results from these experiments should be treated as tentative and preliminary, and further investigations are required on the associations between species composition and soil conditions and subsequently on manipulations of soil conditions. On the basis of results obtained the following conclusions can be drawn:

Each change in site conditions, i.e. moisture content and the content of the available forms of elements, contributes to gradual transformations in the floristic composition of communities and development of variants exhibiting lower species wealth in comparison with the typical variant.

The highest floristic diversity is obtained by variants of communities of the *Molinio-Arrhenatheretea* class which occur in sites characterized by moderate moisture content where the value of the Ellenberg's indices is as follows: F – from 4 to 6; soil total nitrogen content from 3.9 to 5.9. In such conditions, and on the basis of laboratory analyses, the following values were determined: higher content of the available forms of nitrogen (N-NO<sub>3</sub> at the level of 200 kg ha<sup>-1</sup> and N-NH<sub>4</sub> in the amount of 100 kg ha<sup>-1</sup>); potassium from 3 to 4 mg 100g<sup>-1</sup> soil; magnesium, about 50 mg 100 g<sup>-1</sup> soil, and sodium about 20 mg 100 g<sup>-1</sup> soil.

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# Effects of grassland restoration on plant species richness in Swedish agricultural landscapes

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## Abstract

Reduced management of semi-natural grasslands, leading to encroachment of trees and shrubs, is considered to be one of the main causes of declining biodiversity in agricultural areas in Scandinavia. To counteract this, restoration of grasslands has increased during the last decades. This study shows that plant species richness increases with time since restoration, and that ten years after restoration the plant community has already partly recovered. A complete clear-cut of trees and shrubs, however, does not create favourable conditions for many grassland species, and creating a mosaic by leaving some trees and shrubs will protect plants from heavy grazing and trampling. A time-lag in response of plant diversity to landscape structure has also been found. When analysing restored semi-natural grasslands in Sweden, species diversity was not related to current grassland connectivity of a restored site, whereas high historical grassland connectivity had a positive effect on present species richness. Both current and historical landscape configuration, together with local aspects, should be considered in restoration planning. In the short-term, local factors such as light, nutrient levels and management are important, and in the long-term the maintenance or build-up of grassland plant communities could be enhanced by stepping-stone habitats and increased connectivity.

Keywords: biodiversity, landscape, management, plants, restoration

## Introduction

Land use change during the last century has completely altered the traditional rural landscape in Scandinavia. Small-scale farming has been replaced by intensified and specialized agriculture; but an even larger problem is the negative effect of land abandonment on species diversity (Eriksson *et al.*, 2002). Most of the agricultural biodiversity is found in traditionally managed semi-natural grasslands, and a prerequisite for keeping this richness is to continue managing by grazing or mowing. As these species-rich habitats have decreased both in size and number, the remaining habitats suffer from deterioration and fragmentation. To counteract the negative effect of abandonment and prevent local species extinctions, restoration of grassland habitats has increased during the last decades. Restoration of abandoned grasslands is often carried out by cutting trees and shrubs, and by the use of fire; followed by reinstating livestock grazing or mowing. It is also common that new grasslands are created on former arable fields.

## Materials and methods

We conducted a survey of 30 semi-natural grasslands in south-eastern Sweden, which have been restored during the last decade. We examined the effects of local and regional factors on species richness and composition. The vascular plants at each site were investigated in ten 1-m<sup>2</sup> plots that were randomly distributed. The number of trees and shrubs were counted within a circle with a diameter of 40 meters, randomly chosen at each restored site. The local

restoration factors investigated were: the size of the restored site, the time between abandonment of grazing and restoration, the time elapsed since restoration, and the abundance of trees and shrubs at the restored site. We also investigated the landscape surrounding each of the restored sites. For each site, three maps representing different time layers were analysed: cadastral maps of the present-day landscape, the landscape 50 years ago, and land-cover maps from the beginning of the 20<sup>th</sup> Century. The landscapes were analysed with regard to target site connectivity defined by circles with 2 km, respectively, from the centre of each target site. Connectivity of each target site was examined by analysing the extent of semi-natural grasslands there were within each circle, defined as:  $\sum \exp(-\alpha d_j) A_j$ , where  $A_j$  is the area of the  $j$ :th grassland located at a distance  $d_j$  from the target site, for  $j = 1$  to  $k$ .

## Results and discussion

### *Local perspective*

Of the five local factors studied, two were related to species richness: time elapsed since restoration and the abundance of trees and shrubs. The time elapsed since restoration was positively associated with total species richness at restored sites (linear regression,  $P < 0.05$ ,  $b = 0.41$ ), which is congruent with the findings of other restoration studies (e.g. Tikka *et al.*, 2001). However, traditional species composition, including rare short-lived species, takes longer time to restore. The abundance of trees and shrubs at restored sites was also positively associated with total species richness (Figure 1). Thus, leaving a few trees and shrubs to create a mosaic of different successional stages, where a range of more or less grazing-tolerant species may co-occur, will protect plants from heavy grazing and trampling (Pihlgren, 2007). Similar results were obtained by Söderström *et al.* (2001), who found a positive relationship between trees and shrubs and plant species richness, and an even more positive relationship to diversity of birds and insects. To increase the establishment of grassland species and recreating typical grassland communities, seed-sowing is frequently discussed as a conservation tool (Walker *et al.*, 2004; Lindborg, 2006). Sowing seeds from populations within the same regional species pool after the grazing season is one plausible restoration measure.

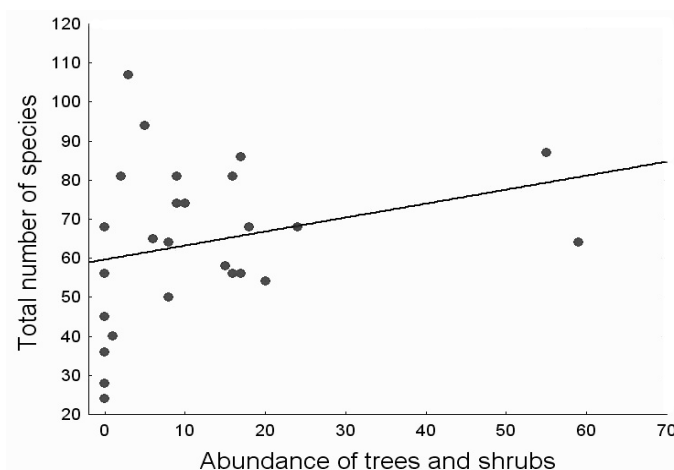


Figure 1. The total number of plant species per site in relation to the abundance of trees and shrubs at restored Swedish semi-natural grasslands (linear regression,  $P < 0.01$ ,  $b = 0.27$ ).

### *Landscape perspective*

Previous empirical studies (e.g. Bruun, 2000) suggest that both area and connectivity of local sites explain a large portion of the variation in plant species diversity among sites. However, none of these factors, when estimated from the present-day landscape, were related to species richness in this study. In contrast, the landscape from the 1950s and the late 19<sup>th</sup> Century

affected variation in present-day plant species diversity, where high historical connectivity was positively related to current species richness (Figure 2). These results indicate that the source areas for diaspores deposited at the target areas have declined over the last century due to drastic land use change during the last 50-100 years. If the build-up of local species richness is a legacy of historically higher connectivity, the lack of relationship between plant species diversity and landscape configuration in the present-day landscape suggests that future loss of species locally may not be compensated for by new colonizations. Hence, the long-term positive effect of restoration will be better enhanced in landscapes with high, than with low, historical connectivity among grasslands.

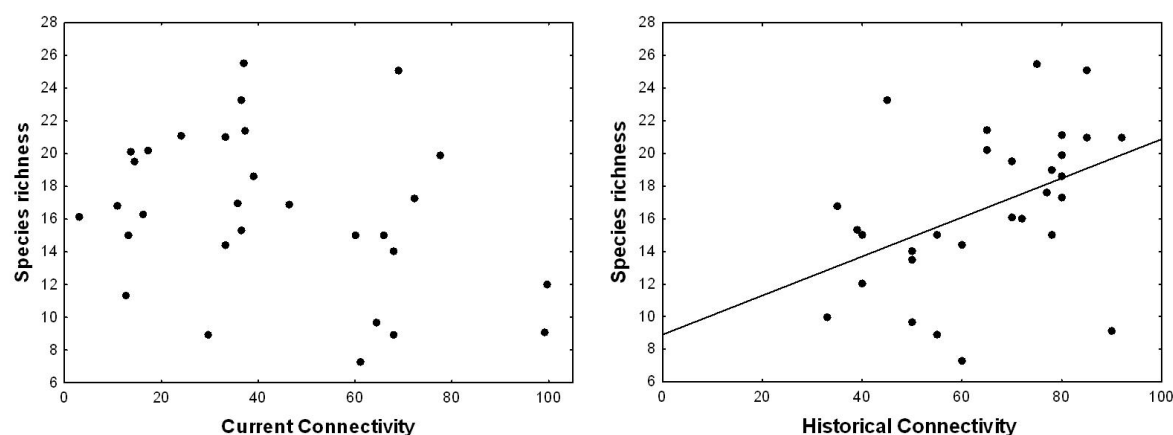


Figure 2. The relationships between species richness and current ( $P > 0.05$ ) and historical (100 years ago) ( $P < 0.01$ ,  $r^2 = 0.52$ ) grassland connectivity in Swedish semi-natural grasslands (linear regression).

## Conclusions

Both local and regional processes are important to consider in grassland restoration. In the short-term, it is crucial to consider local factors such as light and management, to succeed in restoration. However, to maintain or build up grassland plant communities over a long-term perspective and to enhance dispersal, it is not enough to focus on local habitat quality only. Current conservation and restoration efforts are primarily focused on single species-rich grasslands, without considering the effects of the surrounding landscape. If both current and historical landscape configurations, together with local aspects are considered in restoration planning, maintenance of grassland species diversity at a larger landscape scale may also be enhanced.

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# **Biodiversity and ecosystem services in mountain grasslands: a case study in the Planes de Son (Central Pyrenees)**

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## **Abstract**

Models developed for altitudinal grasslands in the Pyrenees show that plant diversity is very sensitive to climatic variables, and that the responsiveness to management factors depends on the particular component representing biodiversity. On the other hand, models developed in the same area for one of the ecosystem services that grasslands provide, soil carbon storage (SOCS), indicate a dependence on both climate and management variables. Variation in vegetation composition, SOCS, and various variables representing different aspects of plant and arthropod diversity, was analysed along an altitudinal gradient in various differently managed ecosystems associated with extensive livestock production in a valley in the Pyrenees. The valley is representative of the traditional grassland management, but has also experienced changes according to the new trends associated with modern livestock production in mountain regions, such as abandonment of marginal areas. Vegetation composition was more responsive to management than diversity, but the interaction between management and altitude complicated the analysis. Grasslands abandoned for a long period and colonized by ash trees showed lower SOCS than grasslands and meadows currently used.

**Keywords:** arthropod diversity, climatic gradients, livestock management, plant diversity, soil carbon storage

## **Introduction**

Land use and climate change are the most important drivers of biodiversity at a global scale (Sala *et al.*, 2000). Ecosystem functions are expected to change with changes in diversity and have become a target of study given the extent of current global changes (Hooper, 2005). Land use and management changes have been significant in the Pyrenees in recent years, associated to a decrease in the rural population. The consequences have been the abandonment of many cut meadows in favour of grazing, a decrease in grazing pressure in marginal areas with the subsequent establishment of forests, and often a change from sheep to cattle-grazing (Sebastià *et al.*, 2008). In order to assess changes in structural and functional properties of grassland along environmental and management gradients, we surveyed grassland communities in a valley in the Central Pyrenees. The valley has a wide altitudinal range (1400-2670 m), and presents a mosaic of management types. Among the structural components we studied plant and beetle species richness and composition, while soil carbon content (SOC) was the studied functional component.

## **Materials and methods**

We surveyed four montane agro-pastoral systems in the valley included in the Planes de Son state (private reserve from the Fundació Territori i Paisatge): 1) cut meadows; 2) abandoned meadows, currently grazed; 3) abandoned meadows, currently occupied by a young ash forest; and 4) cattle-grazed grasslands. In the subalpine and alpine belts, above 1,800 m, we studied grasslands grazed by either cattle or sheep. Plant inventories were made in 100 m<sup>2</sup>

plots. Arthropods were sampled in the six agro-pastoral systems studied, each replicated twice. Three pitfall traps were set at each site, within the 100 m<sup>2</sup> plot. All agro-ecosystems in the montane belt were sampled for two weeks (in June-July). Additionally, all pastured grasslands were sampled for a week at the end of August.

## Results

Both management and altitude influenced plant composition, as shown by the ordination along the first axis of a detrended canonical analysis (DCA, Figure 1). In a restricted canonical analysis the variance explained by these two factors accounted for 39%. Nevertheless, variance partitioning showed that the altitudinal gradient accounted for 10% and management for 34%, indicating a 5% interaction. Plant species richness was lower for forests ( $P = 0.0038$ ) and cut meadows, and higher for montane grazed grasslands (Figure 1). In contrast, carabid beetle species-richness reached the highest values in the forested ecosystems, although not significantly different with the data available ( $P = 0.233$ ). Carabid composition was also affected by management and altitude (Figure 2). SOCS were higher in cut meadows and alpine grasslands and minimal in the young ash forests, although further data are needed to investigate this tendency ( $P = 0.113$ ).

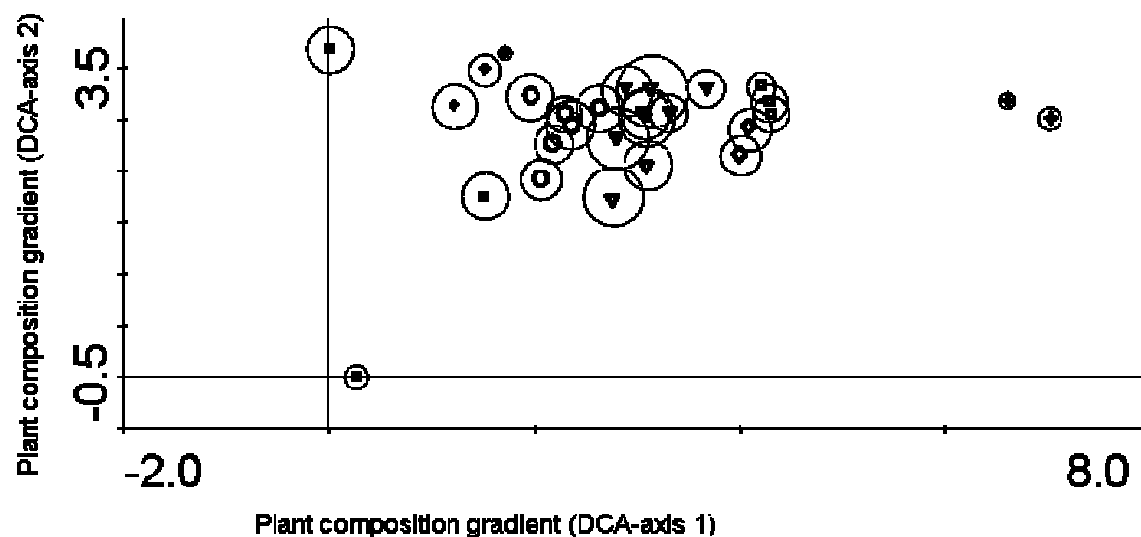


Figure 1. Plant composition gradient represented by the first and second axis of a detrended canonical analysis (DCA). Montane cut meadows (□), abandoned cut meadows currently grazed (◇), and totally abandoned meadows holding a young forest (◆), montane pastures (▼), subalpine cattle grazed pastures (○), subalpine sheep and cattle grazed pastures (◆), alpine sheep grazed pastures (■). Circle size indicates plant species richness.

## Discussion

Climate variation associated with the altitudinal gradient and management mosaic drove plant composition and richness (Figure 1). Although plant diversity may be enhanced in pastures, as compared with cut systems and forests derived from abandonment of all agro-pastoral activities, the diversity of some groups in the consumer trophic level might be higher in abandoned systems such as forests. Different responses to climate and management have been

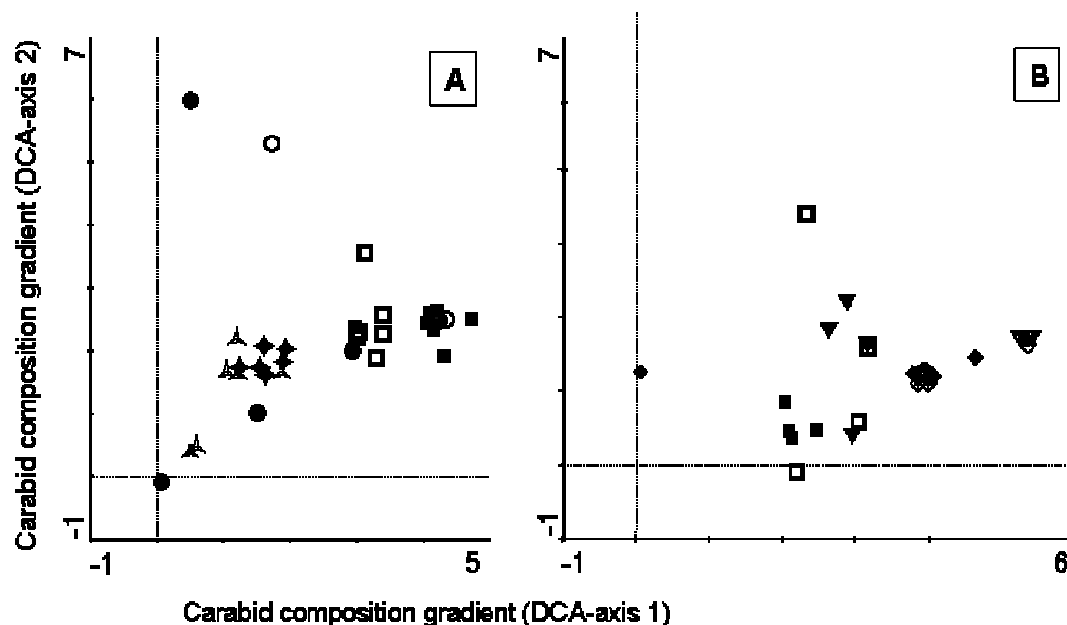


Figure 2. Composition changes in carabid beetle communities along the first and second axes of a detrended canonical analysis (DCA) for A) June-July sampling in montane cut meadows (●, ○), abandoned cut meadows currently grazed (■, □), and totally abandoned meadows holding a young forest (▲, ◆). B) August sampling in montane and subalpine pastures: abandoned cut meadows, currently grazed (■, □), montane pastures (▼, ▽), subalpine cattle grazed pastures (◆, ◇), subalpine cattle and sheep grazed pastures (◆, ◇).

found in different diversity indices (de Bello *et al.*, 2007). The C response tendency found could be associated with intensification including high fertilizer inputs in cut meadows (Conant, 2003). Land use changes derived from socio-economic changes, such as those which have prompted the abandonment of all agro-pastoral practices, may result in major changes in structural and functional variables, as reflected by the change in plant community composition in established post-abandonment forests. On the other hand, changes in composition between differently managed systems reflect the importance of the existence of a variety of management strategies for maintaining  $\beta$ - and  $\gamma$ -diversity at the landscape level.

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# Evolution of different pasture species in a silvopastoral system of NW Spain

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## Abstract

The objective of this experiment was to test the effect of sewage sludge, potassium (K) and NPK fertilization on different pasture species developed under *Pinus radiata* during three years. The experiment was located in Lugo (NW Spain) on abandoned agricultural land. In autumn 1997, a pasture mixture was sown (*Lolium perenne* L., *Dactylis glomerata* L. and *Trifolium repens* L.) and in winter 1997, a *Pinus radiata* D. Don plantation was established (density 1667 trees ha<sup>-1</sup>) over the pasture. Eight treatments were applied: three rates of sewage sludge, each with or without K; and two control treatments, no fertilization (NF) and inorganic fertilizer at usual amounts used in the region. Fertilization treatments were applied during three years. In the first two years, K input with the lowest sewage sludge dose maintained a high clover proportion. However, proportion of sown grasses was increased with nitrogen fertilization. *Dactylis glomerata* was the species with the best establishment and persistence in time.

## Introduction

Agroforestry represent a complex series of ecological interactions to achieve output and environmental benefits by exploiting spatial heterogeneity above and below ground. Tree and pasture competitive relationships depend on pasture response in terms of production and botanical composition, which usually varies with fertilization treatments. Applications of fertilizer to silvopastoral systems have been shown to increase both pasture and tree growth, therefore without harmful effects on trees (Rigueiro *et al.*, 2000; Mosquera *et al.*, 2006). Fertilizer can be applied as inorganic and organic fertilizer but the effects of organic fertilizer on pasture quality and production have been less extensively evaluated than the effects of inorganic fertilizer. Pasture quality and production depends on botanical composition, which in turn is strongly dependent on soil nutrient availability and, therefore, fertilization treatments. Studies on the use of sewage sludge as a fertilizer, which is regulated by EU Directive 86/278/CEE (European Commission, 1986), have concluded that sewage sludge is a good fertilizer due to its high content of macronutrients, particularly N. However, the use of sewage sludge also has to take into account the higher proportion of heavy metals in the sludge than in the soil, as these are toxic elements that can enter the food chain constituting a health risk (Smith, 1996). The objective of the experiment was to evaluate the effect of potassium, sewage sludge and mineral NPK fertilization application on botanical composition.

## Materials and methods

The experiment was carried out in an agricultural land. Soil analyses revealed a pH (water) of 6.3, high nitrogen (2.3 g kg<sup>-1</sup>) concentrations, available Olsen-phosphorus (0.03 g kg<sup>-1</sup>) and medium level of available potassium (K<sub>2</sub>O: 0.09 g kg<sup>-1</sup>). The experimental design was a completely randomized block with three replicates and eight treatments, therefore 24 plots in total. Pasture was established in autumn 1997 before tree planting, when each plot was sown with a mixture of 25 kg ha<sup>-1</sup> of *Lolium perenne* cv. Brigantia, 10 kg ha<sup>-1</sup> of *Dactylis glomerata*



cv. Artabro and 4 kg ha<sup>-1</sup> of *Trifolium repens* cv. Huia after ploughing. In January 1998, a plantation of *Pinus radiata* D. Don was established at a density of 1667 trees ha<sup>-1</sup>. Each plot was constituted by a perfect square of 5 x 5 trees with an area of 96 m<sup>2</sup>. Eight treatments were established and consisted of three annual anaerobic digested sewage sludge doses (S1: 160 kg N total ha<sup>-1</sup>; S2: 320 kg N total ha<sup>-1</sup>; S3: 480 kg N total ha<sup>-1</sup>), with or without potassium (200 kg K<sub>2</sub>O ha<sup>-1</sup> per year) in each case. Two control treatments were also included in the comparison: no fertilizer (NF) and inorganic fertilizer as usually used in the region (MIN: 80 kg N ha<sup>-1</sup>, 120 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and 200 kg K<sub>2</sub>O ha<sup>-1</sup>). All fertilization treatments were homogeneously applied in the spring of 1998, 1999 and 2000. In total, twelve pasture harvests took place in May, June, July and November 1998 and 2000, and April, May, July and December in 1999. Two replicates per plot (0.09 m<sup>2</sup>) were taken by using a hand clipper. Botanical composition was estimated by hand separation, and samples were dried (80°C for 72 hours). Data were analysed using ANOVA with treatments and years as factors. Means were separated by the Duncan test when ANOVA was significant.

## Results and discussion

In general, the sown species proportion was high (Figure 1), because of the initial high soil fertility that gave advantage to sown species. The other species found were grasses *Holcus lanatus* L., *Poa* spp, *Agrostis* spp, *Arrhenatherum elatius* (L.) P.Beauv. ex J.Presl&C.Presl, *Lolium multiflorum* Lam), and dicotyledoneous species (*Rumex obtusifolius* L., *Plantago lanceolata* L., *Erodium moschatum* (L.) L'Hér. and *Taraxacum officinale* Weber, but their contributions were very low (< 150 g kg<sup>-1</sup> DM harvest in most cases) and similar in the different treatments.

The use of potassium, when the sewage sludge dose was low, improved the clover contribution. In 1998 and 1999, the highest clover proportion was observed with no fertilization treatment (680 and 370 g kg<sup>-1</sup>, respectively). Similar values were obtained when the lowest sewage sludge dose (S1) with potassium fertilization (200 kg K<sub>2</sub>O ha<sup>-1</sup>) was applied (370 and 330 g kg<sup>-1</sup>, respectively). By contrast, in 1999, the *L. perenne* proportion was increased with nitrogen fertilization (sewage sludge and mineral NPK treatments) (200-390 and 320 g kg<sup>-1</sup>, respectively, compared to 60 g kg<sup>-1</sup> with no fertilization). A negative effect of nitrogen fertilization on legume proportion, which implies an advantage for grass species, was detected (Whitehead, 1995). In this area, González (1992) observed that grass proportion was increased with the application of N over 50 kg N ha<sup>-1</sup> year<sup>-1</sup> (that is higher than N applied with S1), but clover was significantly reduced with more than 100 kg N ha<sup>-1</sup> year<sup>-1</sup>; that explains the nil response to potassium application with S2 and S3. An incremental increase in sown grass proportion with sewage sludge dose was observed by Rigueiro-Rodríguez *et al.* (2005) in a silvopastoral system located on an acid soil very poor of nutrients. However, in the Rigueiro *et al.* experiment, the increment of sown grasses was higher (between 200-320%) compared with no fertilization, because, when no fertilization was applied, the presence of sown species (better adapted to fertile conditions) was much lower (5-19%) in the first year. In June 2000, maximum sown grasses were observed with mineral fertilizer (760 g kg<sup>-1</sup> compared to 590 g kg<sup>-1</sup> with NF), of which 710 g kg<sup>-1</sup> of the total pasture proportion was *D. glomerata* and only 50 g kg<sup>-1</sup> was *L. perenne*. Differences between treatments were few, due to the low proportion of *L. perenne* in 2000. However, sown species proportion was high, between 600-900 g kg<sup>-1</sup> due to *D. glomerata* contribution in all treatments. In the no-fertilization treatment, N contribution of legume to soil, which is rich in nitrogen (Whitehead, 1995) could enhance the good development of this grass. It is important to highlight that *L. perenne* is a species that is established quickly, but its persistence in time is usually very low in our region, where summer drought reduces its proportion. However,

*D. glomerata* was the species with the best establishment and persistence in time. Then, it is very adequate for silvopastoral systems (Mosquera *et al.*, 2001).

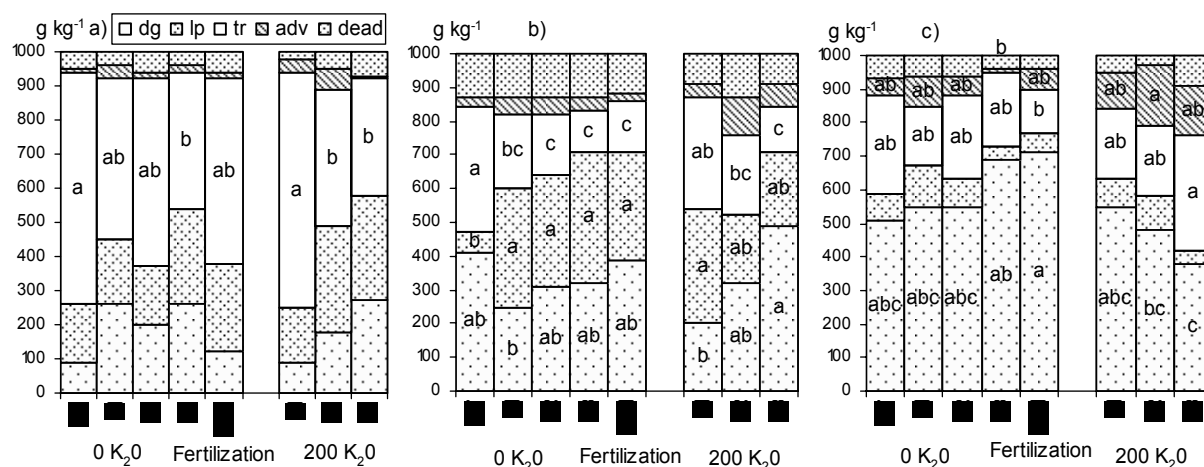


Figure 1. Proportion in weight ( $\text{g kg}^{-1}$  dry matter) of *Dactylis glomerata* (dg), *Lolium perenne* (lp), *Trifolium repens* (tr), adventitious species (adv) and dead matter (dead) in pasture under different fertilizer treatments in a) June 1998, b) July 1999 and c) June 2000. NF: no fertilization; S1: low sewage sludge ( $160 \text{ kg total N ha}^{-1}$ ); S2: medium sludge ( $320 \text{ kg total N ha}^{-1}$ ); S3: high sludge dose ( $360 \text{ kg total N ha}^{-1}$ ); MIN: mineral fertilization ( $80 \text{ kg N ha}^{-1}$ ,  $120 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$  and  $200 \text{ kg K}_2\text{O ha}^{-1}$ ). Different letters indicate significant differences between fertilization treatments ( $P < 0.05$ ).

## Conclusions

Potassium and the lowest sewage sludge dose combination maintained high clover proportion during the first years of establishment. However, the proportion of sown grasses, mainly *Dactylis glomerata*, was increased with nitrogen fertilization as the sward developed.

## Acknowledgment

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# The effects of different kinds of livestock farming and abandonment on botanical diversity in mountain hay meadows

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## Abstract

The process of agricultural abandonment has proceeded at an alarming rate during recent decades in many European mountain areas. The process has been even greater for traditional livestock farming, as it is in the case of the extensive management that is undertaken in the Catalan Pyrenees. Research is conducted here on the importance of the traditional livestock farming for botanical diversity conservation in mountain hay meadows (*Arrhenatherium elatioris*) in the county of Pallars Sobirà in the Catalan Pyrenees (Spain). The aim is to analyse the effects on the botanical diversity of these meadows of the changing practices prompted by the process of abandonment. A comparison was carried out among hay meadows grazed by sheep, horses and cattle, and also between cut meadows and partially abandoned ones. Botanical transects have been carried out by means of the point intercept method. Transects have been analysed using different biodiversity indices in order to consider both species abundance and species richness. Finally, our study reveals the existence of a small tendency in the process of agricultural abandonment to undermine the characteristic botanical composition of mountain hay meadows.

Keywords: mountain hay meadow, agricultural abandonment, biodiversity, extensive livestock farming

## Introduction

In the Catalan Pyrenees, as in the great majority of the European mountain regions, agriculture is declining (MacDonald *et al.*, 2000). This process is particularly severe in the case of traditional agriculture. This course of action is of even greater concern when considering the goods and services, apart from the usual farm produce, that this kind of agriculture provides for society. The role of agriculture in preserving biodiversity is among the most valued alternative functions of traditional mountain agriculture. Thus, we performed an investigation in a region of the Catalan Pyrenees, in the county of Pallars Sobirà, on how the process of agricultural abandonment affects botanical diversity in mountain hay meadows. Agriculture in this area is characterized by traditional livestock farming, which consists of an extensive management of the herd – sheep, cattle and horses – between the alpine grassland in summer and the hay meadows of the middle-altitude lands in winter. The traditional management of hay meadows comprises a cutting in summer for hay forage production, to be fed in winter, and grazing of the regrowth in autumn. The process of agricultural abandonment in the study area is being characterized mainly by two processes: (a) the substitution of mowing by additional grazing, that is, the shift from cut meadows to partially abandoned meadows; and (b) while sheep farming is declining and cattle farming keeps relatively stable, horse farming is increasing. The aim of this study is to analyse the effects on the botanical diversity of mountain hay meadows, of the effects of different meadow managements and livestock farming.

## Material and methods

A total of 156 transects were recorded in 20 different mountain hay meadows between 2005 and 2006. Each hay meadow was sampled twice, once in summer – before the mowing or grazing – and again in autumn – before the grazing. All meadows shared similar features, namely: unirrigated land; at least 20 years of fidelity to the same meadow management and livestock farming; between 1,100 and 1,400 metres above sea level; west aspect; and all of them had been cornfields in the past. Plant cover was estimated using the point intercept method, as described by Sebastià (1991) for hay meadows in the Catalan Pyrenees. Each sampling consisted of four 5-metre lineal transects laid out in a randomized block design. All the species intercepted by a vertical pointer at 10 cm intervals were recorded along these lines. In order to take into account both components of biodiversity measurement, abundance and evenness, species diversity was calculated using four different indices, namely: Species Richness (S), Simpson index (D), Shannon index (H') and Equitability index (E). Species abundance is the number of different species found in a certain ecosystem. Species evenness quantifies how equally represented are the species of a given community (Magurran, 1988). Thus, an ecosystem where all the species are found by the same number of individuals would have high species evenness. While Species Richness only considers species abundance, and Equitability index explains merely species evenness, Shannon and Simpson indices account for both. The effects of differential meadow managements and livestock farming on these two components of biodiversity measurements were tested by one-way ANOVA, given that the variables were found to follow a normal distribution. When statistically significant effects were identified, the ANOVA *post-hoc* Fisher's PLSD test ( $P < 0.05$ ) was also applied.

## Results and discussion

Management does not seem to have a strong influence on the botanical diversity of mountain hay meadows. No difference was observed between cut and partially abandoned meadows regarding species evenness, since no significant differences in the botanical composition were detected between cut and partially abandoned meadows, as shown by the results of Simpson, Shannon and Equitability indices (Table 1). The only biodiversity index that show significant differences between cut and partially abandoned meadows is Species Richness. More species abundance was detected in partially abandoned meadows. The absence of mowing seems to promote the presence of species more characteristic of pasture communities – for instance, *Luzula campestris* – which are not so common in the community of mature mountain hay meadows (*Arrhenatherum elatioris*).

Table 1. Effects of meadow management on botanical diversity in mountain hay meadows according to several diversity indices (mean  $\pm$  S.E.).

	Meadow management		Anova results	
	Cut meadow n = 44	Partially abandoned meadow n = 112	F	P
Richness	23.6 $\pm$ 0.46 (a)	25.8 $\pm$ 0.92 (b)	5.81	0.017
Simpson	0.13 $\pm$ 0.003	0.13 $\pm$ 0.006	1.51	0.221
Shannon	2.36 $\pm$ 0.024	2.38 $\pm$ 0.040	0.20	0.655
Equitability	0.75 $\pm$ 0.007	0.74 $\pm$ 0.010	1.36	0.246

Table 2. Effects of livestock farming on botanical diversity in mountain hay meadows according to several diversity indices (mean  $\pm$  S.E.).

	Cattle n = 48	Livestock farming		Anova results	
		Horse n = 48	Sheep n = 60	F	P
Richness	23.8 $\pm$ 1.00	25.2 $\pm$ 0.60	23.7 $\pm$ 0.60	1.20	0.305
Simpson	0.12 $\pm$ 0.005 (a)	0.14 $\pm$ 0.006 (b)	0.12 $\pm$ 0.004 (a)	6.53	0.002
Shannon	2.43 $\pm$ 0.038 (a)	2.30 $\pm$ 0.037 (b)	2.36 $\pm$ 0.029 (ab)	3.17	0.045
Equitability	0.78 $\pm$ 0.007 (a)	0.72 $\pm$ 0.010 (b)	0.75 $\pm$ 0.009 (a)	10.15	< 0.0001

With regard to livestock management, although differences in botanical composition among meadows are again small, they are statistically significant in all biodiversity indices considered except for the Species Richness. Meadows grazed by horses have greater species abundance, while meadows grazed by cattle and sheep show higher species evenness (Table 2).

## Conclusions

Our study reveals that the different meadow management and livestock farming practices considered do not have important effects on the botanical diversity of mountain hay meadows. Nonetheless, significant differences were detected that seem to point to the existence of a tendency which shows that meadows grazed by horses are richer in species abundance, while meadows grazed by cattle and sheep show higher species evenness. Although the differences are not so significant, they are similar as regards the type of meadow management, since partially abandoned meadows show higher magnitudes of species richness, while cut meadows show higher species evenness. Consequently, it can be said that despite the small differences identified, the process of agricultural abandonment is undermining the characteristic botanical community of the mountain hay meadows, as shown by the advent of more ruderal species and species more characteristic of pasture communities in the meadows grazed by horses and in the partially abandoned meadows.

## Acknowledgements

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## Analysis of weed diversity in alfalfa (*Medicago sativa*)

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### Abstract

A research programme was initiated in 2005 in a study area in the Deux-Sèvres region in western France to evaluate grassland impacts on biodiversity. One of the objectives deals with the determination of weed diversity in alfalfa and the identification of the agro-ecological factors explaining plant community structure and dynamics. Sixty-nine alfalfa fields were investigated in 2006 and 61 in 2007, and sampling determination of part of the heterogeneity of agronomic and environmental conditions, such as alfalfa age, agricultural management and field environment. Fifteen fields, that were one-year-old in 2006, were observed both years. The same protocol for weed community estimation was followed: 30 stations were observed in each field (distributed on two or three transects, depending on the field geometry). Plant diversity is characterized by classical indicators, such as species richness and abundance, Shannon index and Evenness index. Results show a large effect of grassland management on plant diversity while age effect is less consistent. For the diachronic survey of the 15 fields, effects of age and management are not significant. A change in plant community is observed to the benefit of perennial species.

Keywords: plant diversity, temporary grassland, farmer fields, agro-ecological determinants

### Introduction

Farmland biodiversity has encountered a severe decline for several decades in Europe and North America. Agriculture intensification is now clearly involved (Chamberlain *et al.*, 2000) but, more precisely, it is the decline in heterogeneity it causes in landscapes that seems to be more detrimental (Benton *et al.*, 2003). Grasslands, which represent a large part of the land use in France and elsewhere in Europe, have seen their surfaces dramatically declining during the same period (French agriculture statistical data) to the benefit of more intensive cropping systems (Poschlod *et al.*, 2005). A research programme was initiated in the Poitou-Charentes region to evaluate the role of grasslands in maintaining biodiversity at the regional scale. Grasslands may contribute to habitat quality for biodiversity in several ways: protection, reproduction and trophic resources availability, including for species of conservation concern, such as the little bustard (Wolff *et al.*, 2001). I hypothesized that these ecological functions depend on plant diversity in grasslands. The study focused on alfalfa grasslands, in which common weed diversity may contribute to plant diversity according to the age of grasslands. Weed community and diversity in alfalfa grasslands were studied considering grassland age and management.

### Materials and methods

The study area is located in the Deux-Sèvres region in western France. Sampling of alfalfa (*Medicago sativa*) fields was carried out in order to have different age classes (1 to 5 years) and to cover the entire study area as much as possible. Sixty-nine fields were studied in 2006 and 61 in 2007. Fifteen fields, which were one-year-old in 2006, were observed both years. Weed flora was determined by noting weed species in 0.25 m<sup>2</sup> circles (mark 1 when observed once, mark 2 when observed more than once). Observations were repeated 30 times along two or three transects depending on field geometry, and data positions were monitored with a

standard GPS. Measurements were carried out from mid-April to mid-May. Plant diversity variables, such as species richness (S) and abundance (A), Shannon (H) and evenness (E) indices were computed from weed flora data. Species abundance at field level is calculated as the sum of the cumulative mark of each species. Agronomic data concerning grassland management (N, P and K fertilization, herbicide treatments, date and number of cutting) were collected during farmer interviews. Fields, in which an AEA (Agri-Environmental Agreement) was contracted, were denoted. Agronomic data of 20 fields were obtained in 2006 and 46 in 2007. Analyses of variance were performed with R freeware. Effects of year, age and management were tested. Management was described with a synthetic datum that integrates N, P and K fertilization, weed management and AEA. Six levels were defined: AEA, AEAxW (AEA with weed management), W (weed management only), Fert (fertilization only), FertxW (fertilization and weed management) and Null (no treatments).

## Results and discussion

Management effect is significant for all diversity variables ( $P < 0.01$ ), excepted for E. Year and age effects are significant only for species abundance ( $P = 0.03$  and  $0.01$ , respectively).

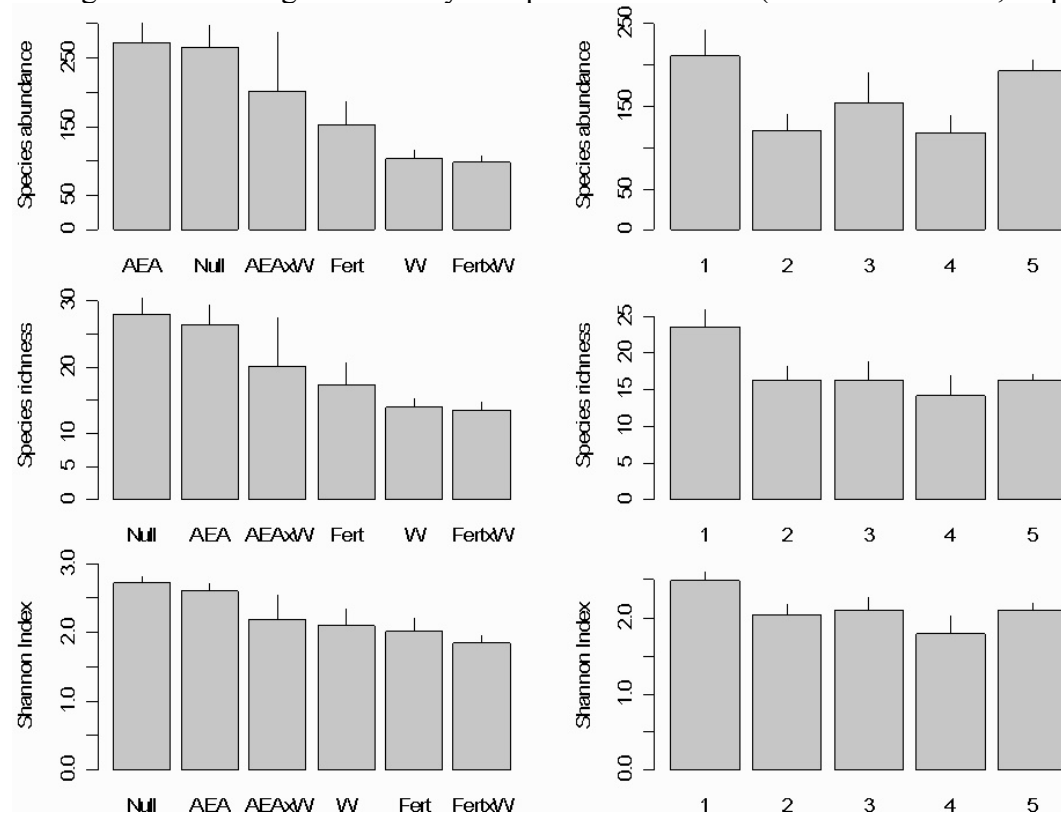


Figure 1. Species abundance and richness and Shannon index vs. grassland management (on the left) and grassland age (on the right). Bars are standard errors.

A, S and H present higher values in extensive management (AEA and Null), where there is no or little use of fertilization and weed management (Figure 1). Fields with weed management (W and FertxW levels) are the least diverse. Plant diversity in fields with AEAxW and Fert management levels is intermediate. For the age effect, there is a large decrease in abundance between years 1 and 2. It remains almost constant in 2- to 4-year-old grasslands before increasing at age 5. This trend is not observed for species richness and Shannon index even though we can notice a slight decrease between years 1 and 2.

There is no significant effect of age and management for the 15 fields surveyed in 2006 and 2007. When considering the most frequent weeds (observed in more than 10% of the circle observations), we observed a modification in the representation of the biological types. In one-year-old grasslands, annual weeds were in the majority (*Veronica persica*, *Capsella bursa-pastoris*, *Atriplex patula*, *Sinapis arvensis*, *Picris echinoides*, for instance) with three biannual species (*Sonchus asper*, *Sonchus oleraceus* and *Petroselinum segetum*). Plants with perennial behaviour increased a lot in two-year-old fields, such as, in order of importance, *Taraxacum officinalis*, *Silene latifolia*, *Poa trivialis*, *Convolvulus arvensis*.

Diversity indices measured in this study are within the range of those reported for similar cropping systems (Légère *et al.*, 2005). Shannon and evenness indices show high values certainly due to the method of estimation of species abundance. This point should be considered for comparison with others studies. Nevertheless, it allowed comparison of weed diversity of fields in our study.

Plant diversity in alfalfa grasslands is mainly determined by the degree of intensification of grassland management. This result is commonly observed in permanent grasslands (Duru *et al.*, 1998) but less in the temporary ones because they are less studied. In this study, AEA management involves less use of fertilization and weed management but also a long period (15<sup>th</sup> May to 31<sup>st</sup> August) without any operation in the field, like cutting. The latter condition is used to increase nesting success of the little bustard, a farmland bird of conservation relevance in the study area. This measure may also permit plant flowering and persistence in fields explaining higher plant diversity encountered in these grasslands.

Weed abundance diminution during the first years of grasslands is probably related to alfalfa competition. Alfalfa productivity declines after several years of cultivation, leaving some space available for weed growth. This may explain the increase in weed abundance in five-year-old grasslands. Modification in plant community is also observed in the first two years. Perennial plants, common in grasslands, seem to become established from the second year of grasslands, to the detriment of annual plants, common in cultures. Further analyses of plant life forms are required at the entire level of the plant community.

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# Ecological and economical technology for mechanization of grassland improvement works

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## Abstract

In the paper are presented new technological solutions for mechanization of improving the grassland working, using the special farming machinery: direct drilling machine, sowing machine and a complex aggregate composed by rotary tiller-drill machine and a fertilizer equipment. These machines were specially designed and made by Grassland Research and Development Institute Brasov-Romania.

The testing results, presented in the second part of the paper, prove the distinct performances of the new technologies, both concerning the agricultural and functional requirements, fuel and work force economy performances.

In comparison with usual technologies, the new technologies involves a lower specific fuel consumption with 8.63-25.4%, a lower work force consumption with 14.63-58.11% and the minimum passing number of the units.

Keywords: grassland improvement, special machines, technological solutions

## Introduction

The degraded status of the considerable grassland area of Romania, about 4.9 m ha, requires improvements or maintenance of the fertile areas with high green mass production by using adequate cultivation technologies. The improvement of degraded pastures by reseeding (total renewing) has an important role, because this can enable an increase of 3-5 times in forage and animal production, while according with biodiversity conservation, the landscape beauty, environmental protection and other advantages in compliance with EU rules. This paper presents the results of technological solutions for improvement of degraded grassland using the reseeding method.

## Materials and methods

The proposed technologies use the new performing equipments and farming machines: the special grass seeders MSPFP 2.5 type (Mocanu *et al.*, 2003), the improved grassland rotary tiller-drill machine MCT 2.5M type (Hermenean *et al.*, 2001), and frontal fertilizing equipment. Besides the current practice these new agricultural machines can be used for achievement of high quality work performances in according with reduced inputs and a low impact on the environment. The trials of the improvement technology for grassland renovation were carried out in the period 2005-2007, in two areas with different conditions: a degraded pasture with deep fertile soil and deep turf, respectively, and a degraded pasture with thin fertile soil and deep turf. The field experiments were carried out both during spring and the summer-end period.

The area conditions of the experiments for degraded grassland with deep fertile soil and deep turf are: type of soil – deep podzols on acid clay dump; humidity of soil at 10-25 cm depth, 25.8%; type of pastures – *Nardus stricta* with *Deschampsia caespitosa*; sown mixture species

*Phleum pratense* 35%, *Festuca pratensis* 25%, *Lolium perenne* 10%, *Trifolium pratense* 20%, *Lotus corniculatus* 10%; seeding rate was 36 kg ha<sup>-1</sup>; and fertilizing rate was N100, P50, K50. The area conditions of the experiments for degraded pastures with thin fertile soil and deep turf are: type of soil – Eutric cambisols on basic sandstone; humidity of soil at 5-12 cm depth, 24.2%; type of pasture – *Agrostis tenuis*; sown mixture species: *Dactylis glomerata* 25%, *Festuca pratensis* 15%, *Festuca arundinacea* 15%, *Lolium perenne* 10%, *Medicago sativa* 15%, *Trifolium pratense* 10%, *Lotus corniculatus* 10%; seeding rate was 38.5 kg ha<sup>-1</sup>; and fertilizing rate was N50, P50, K50.

The conditions of the experiments are presented in Table 1.

Table 1. Conditions of the experiments.

Type of grassland	Technology	Period of seeding	Farming machine <sup>*</sup>
Degraded with deep fertile soil and deep turf	Usual	Spring	u1-u2-u3-u4-u5-u6-u5-u7-u8-u9
		summer –autumn	u1-u2-u10-u3-u4-u5-u6-u5
	New	Spring	u1-u2-u4-u12-u7-u8-u9
		summer –autumn	u1-u2-u13-u12
Degraded with thin fertile soil and deep turf	Usual	Spring	u1-u11-u11-u3-u5-u6-u5-u7-u8-u9
		summer –autumn	u1-u11-u11-u3-u5-u6-u5
	New	Spring	u1-u15-u7-u8-u9
		summer –autumn	u1-u14

<sup>\*</sup> u1-u14 from the Table 1 have following significations: u1- Machine for grassland clearing of mole hills and non value vegetation; u2 – Reversible plough; u3- Centrifugal fertilizer spreader; u4-Total soil cultivation equipment; u5-Ring roller; u6-Universal seed drill; u7- Rotary disc mower; u8- Rotary tractor-rake; u9-Self loading truck; u10- Disc harrow; u11-Rotary tiller; u12- Special grass sowing machine, MSPFP 2.5 type, in aggregate with the frontal carried fertilizer spreader, EF 2.5 type; u13 –Power harrow for seedbed preparing; u14-Rotary tiller-drill machine, MCT 2.5M type in aggregate with the frontal carried fertilizer spreader, EF 2.5 type. Each experimental plot, carried out in different area conditions and seeding period, has 1ha surface.

## Results and discussion

The forage yields during the period 2005-2007 are presented in Table 2. There were eight combinations of types of grassland and seeding periods. From each plot five samples were taken for determining botanical composition and the dry matter yield.

Table 2. Dry matter yields after grassland improvement.

Type of grassland	Technology	Period of seeding	DMY [Mg ha <sup>-1</sup> ]	DMY [Mg ha <sup>-1</sup> ]	DMY [Mg ha <sup>-1</sup> ]
			2005	2006	2007
Degraded with deep fertile soil and deep turf	usual	spring	5.2	8	5.2
		summer-autumn	-	7.96	5.16
	new	spring	5.24	8.12	5.24
		summer-autumn	-	8.1	5.22
Degraded with thin fertile soil and deep turf	usual	spring	4.84	6.82	4.26
		summer-autumn	-	6.68	4.12
	new	spring	4.88	6.9	4.31
		summer-autumn	-	6.74	4.18

Table 2 shows us the DM yields are close for different technologies. The relevant differences consist in specific fuel consumption, number of the unit passes, and the necessary work force, presented in Table 3 and Figure 1. Analysis of these parameters shows the following: the specific fuel consumption is lower by 8.78–25.4% for new technologies in comparison with usual technologies; the work force necessary for new technologies is reduced by 14.63-58.11% in comparison with usual technologies; the number of unit passes is reduced from a maximum of 10 for the usual improvement technology of grassland with deep

fertile soil, during the spring period, up to 2 passes for new improved technology for grassland with thin fertile soil, at the late summer – early autumn period.

Table 3. Results of experiments

Type of grassland	Technology	Period of seeding	Fuel consumption [l ha <sup>-1</sup> ]	Work force necessary [man hours ha <sup>-1</sup> ]	Number of unit passes
Degraded with deep fertile soil and deep turf	usual	spring	98.65	12.37	10
		summer –autumn	66.5	9.11	8
	new	spring	89.25	10.56	7
		summer –autumn	60.8	6.76	4
Degraded with thin fertile soil and deep turf	usual	spring	99.4	14.07	10
		summer –autumn	63	10.24	7
	new	spring	83.4	8.12	5
		summer –autumn	47	4.29	2

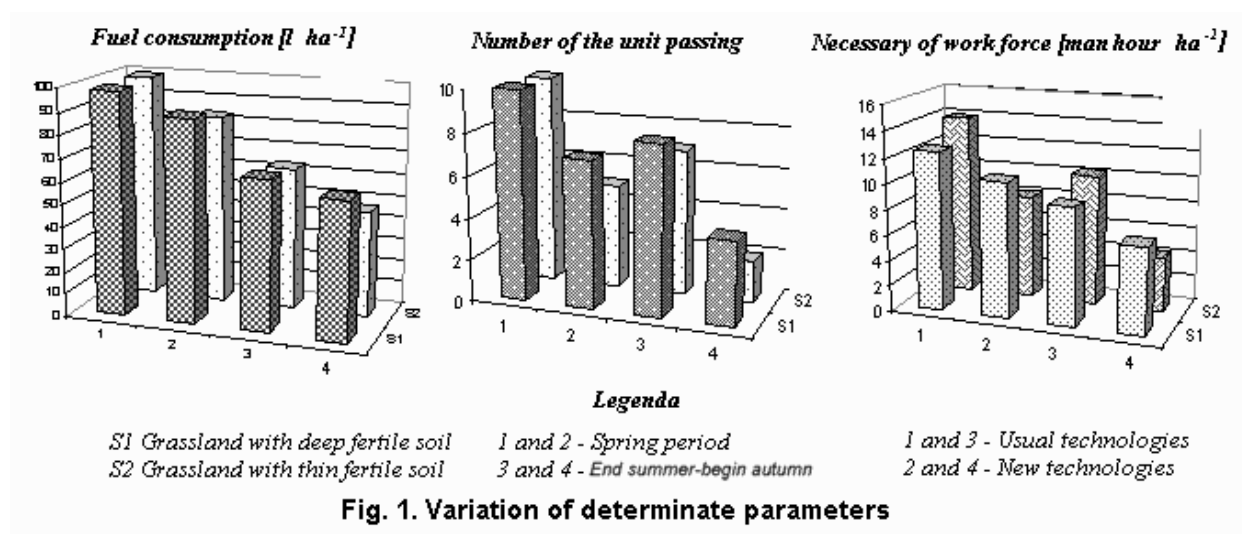


Fig. 1. Variation of determinate parameters

## Conclusions

According to comparative trials of usual and new technologies for grassland improvement the results lead to the following conclusions:

- the forage yields following improvement using the new technologies were similar to the yields following improvement using usual technologies;
- promoting new grassland farming equipments and machines, e.g. special grass seeding machine MSPFP 2.5 type, rotary tiller-drill improved machine MCT 2.5M type, and frontal fertilizer equipment EF 2.5 type, the new technologies are more economical and efficient than usual technologies, because they can reduce fuel consumption by 8.63-25.4%, work force consumption by 14.63-58.11%, and the number of passes of the units (from 10 maximum up to 2 minimum);
- the new technologies of grassland improvement can be successfully applied to realize a good combination of yield, forage quality, botanical composition with cost requirements, according the conditions of the area.

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# Temporal distribution of cattle and horses grazing on mountain rangelands partially invaded by *Euphorbia polygalifolia*

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## Abstract

Horse stocking rates have increased significantly in the last decades in many summer rangelands of northern Spain traditionally used by cattle. Additionally, a major type of grassland within these rangelands (*Festuca-Agrostis* dominated grassland) is experiencing a noticeable invasion by the spurge species *Euphorbia polygalifolia*. The objective of this study was to compare diurnal grazing distribution of cattle and horses in order to provide information on the relationship between horse grazing patterns and pasture degradation.

Temporal distribution of daylight grazing was quantified in eighteen cows and fifteen mares, from June to September 2004, in a representative summer rangeland of northern Spain. Mares spent more diurnal time grazing than cows (81.7 vs. 63.1%;  $P < 0.001$ ). While cows showed two distinct grazing peaks, at early morning and late evening, mares had a more uniform daily grazing pattern. Finally, mares spent a higher proportion of their grazing time on grasslands prone to spurge invasion than cows (97.2 vs. 77.9;  $P = 0.001$ ).

These results suggest that horses could contribute to grassland overgrazing in this type of European mountain rangelands, favouring in this way spurge invasion.

Keywords: overgrazing, grazing behaviour, communal land

## Introduction

Traditionally used by beef cattle, communal summer rangelands of the Atlantic mountains of northern Spain have, in recent decades, experienced an increasing presence of horses. A hypothetical different grazing utilization of the vegetation mosaic by mares and cows may contribute to explain some current undesirable vegetation changes, like the colonization of the spurge species *Euphorbia polygalifolia* on acidic grasslands. High forage utilization of the herbaceous species of this grassland community and rejection of *E. polygalifolia* by both animal species favour spurge abundance (Busqué *et al.*, 2003). The objective of this study was to compare diurnal grazing distribution of cows and mares in a representative upland rangeland, in order to quantify the responsibility of these livestock species in the process of spurge invasion.

## Materials and methods

The experiment was carried out on an acidic communal summer rangeland in the north of Spain (Puerto de Sejos, Cantabria) at a mean altitude of 1700 m. Vegetation is a complex mixture of different plant communities characteristic of the Atlantic region: grasslands dominated by perennial herbaceous species (*Agrostis capillaris*, *Festuca nigrescens*, *Nardus stricta*) dwarf shrubs (*Calluna vulgaris*, *Erica* spp., *Vaccinium myrtillus*) and broomfields of *Genista florida*. The area is included within the European Natura 2000 network.

Livestock grazing behaviour was measured by sampling focal animals continuously (Altmann, 1974) in 33 diurnal sessions (18 cows and 15 mares) grouped in three periods of the grazing season of 2004: 'initial' from 16 June to 15 July; 'intermediate' from 16 July to 15 August; and 'final' from 16 August to 15 September. Every 20 minutes, from dawn to dusk,

the activity (grazing, resting or walking), geographical position and type of vegetation consumed of a single animal was recorded.

Statistical analyses were performed using general linear models. In the case of livestock daily activities, the model included the fixed factors Species and Period and their interaction. For the analysis of grazing activity during the day, the model considered each diurnal hour as a level of a fixed factor. *Post hoc* multiple comparison tests (Tukey method) were used to identify different means. Finally, due to lack of homogeneity of variance, non-parametric analyses were performed for the explanatory variable 'proportion of grazing time spent on grasslands'. All the analyses were done using the software SPSS (SPSS, 1999).

## Results and discussion

In agreement with previous studies (Arnold and Dudzinski, 1978; Menard *et al.*, 2002), mares spent more time grazing than cattle (11.5 vs. 9.1 hours and 81.7 vs. 63.1% of daylight time respectively;  $P < 0.001$ ; Table 1). On the contrary, resting time was lower for mares (2.0 vs. 4.5 hours and 14.2 vs. 31.4% for mares and cows respectively;  $P < 0.001$ ). There was no significant difference in walking time between the two species. Neither a significant effect of the Period nor of the interaction 'Species  $\times$  Period' was observed for any activity.

Table 1. Temporal distribution of daylight activities during summer grazing.

	Species				Period					Species x Period
	Cattle	Horses	SED	<i>P</i>	Initial	Inter.	Final	SED	<i>P</i>	<i>P</i>
Grazing time (h)	9.1	11.5	0.53	< 0.001	10.9	10.7	9.3	0.65	0.043	0.155
Grazing time proportion (%)	63.1	81.7	3.67	< 0.001	70.3	73.6	73.2	4.50	0.715	0.356
Resting time (h)	4.5	2.0	0.48	< 0.001	3.9	2.3	2.7	0.59	0.151	0.310
Resting time proportion (%)	31.4	14.1	3.20	< 0.001	24.9	22.8	20.5	3.90	0.595	0.469
Walking time (h)	0.8	0.6	0.19	0.259	0.7	0.5	0.8	0.24	0.519	0.503
Walking time proportion (%)	5.5	4.2	1.40	0.361	4.8	3.6	6.2	0.02	0.346	0.441

SED: standard error of the difference; *P*: significance level.

Cows showed two noticeable grazing peaks during the day (Figure 1): early in the morning and late in the evening. Grazing activity decreased at mid-day, although it did not disappear completely. This pattern was not so evident in mares, where grazing was the predominant activity even at mid-day.

With respect to the proportion of grazing time spent on grasslands, as opposed to shrub dominated communities, mares showed significant higher values than cows (97.2 vs. 77.9%;  $P = 0.001$ ; Table 2). No differences on this variable were observed due to the period of the season.

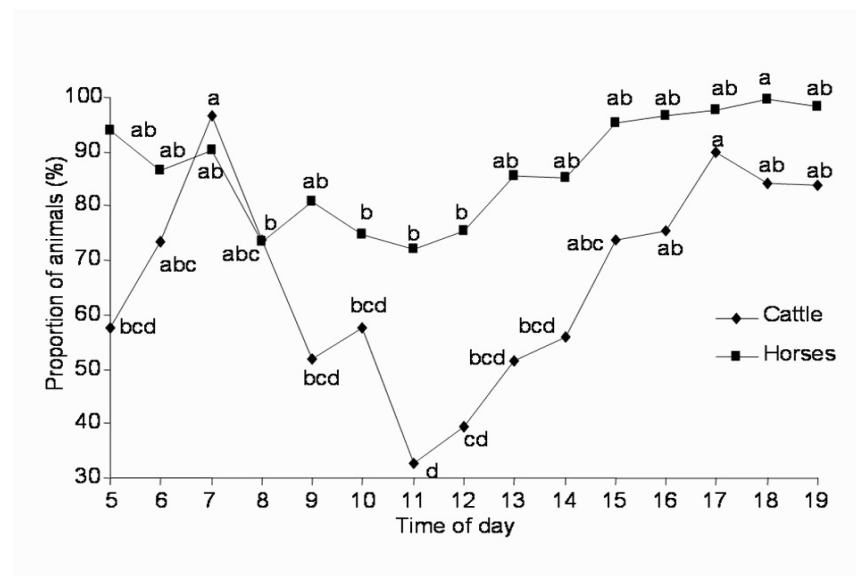


Figure 1. Grazing diurnal pattern of cattle and horses. The y axis presents retransformed percentages (transformation:  $x' = \arcsin\sqrt{x}$ ). Different letters for each species indicate statistically different mean values (Tukey's test with  $P < 0.05$ ).

Table 2. Effect of the species (cattle vs. horses) and the period of the season (initial; intermediate and final) on the proportion of grazing time spent on grasslands (PGTG).

		PGTG (%)	Significance level ( <i>P</i> )
Species	Cattle	77.9	0.001 <sup>1</sup>
	Horses	97.2	
Period	Initial	91.5	0.311 <sup>2</sup>
	Intermediate	79.9	
	Final	88.4	

<sup>1</sup>Mann-Whitney test ( $U = 46.0$ ). <sup>2</sup>Kruskal-Wallis test (Chi-Square = 2.3; 2 degrees of freedom).

## Conclusions

Overgrazing of grassland communities in Atlantic upland rangelands is enhanced by the increasing presence of horses, as they use this resource very intensively (58% more than cattle as a daily average) and consistently (without noticeable changes along the grazing season). Sustainable grazing management in this type of ecosystems of high conservation value should consider explicitly the differences in grazing utilization among different herbivores in order to avoid degradation processes as that of the spurge colonization in acidic grasslands.

## Acknowledgements

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# Species rich limestone grasslands of the Burren, Ireland: feed value and sustainable grazing systems

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## Abstract

Winter grazing of the limestone grasslands, pavements and heaths of the Burren is a key factor in maintaining their biodiversity. Over the last 30 years farmers have moved away from store cattle to the grazing of suckler cows. The higher nutritional requirements of in-calf cows resulted in many farmers introducing silage onto winter grazed areas (winterage) or housing animals. This led to reduced grazing on winterages and to farm polarization (intensification and reseeding of improved agricultural grassland, while abandoning semi-natural grassland on the same farm). An EU LIFE Nature funded project 'BurrenLIFE', is developing a new model for sustainable agriculture in the Burren. Central to this is the introduction of grazing regimes which seek to optimize utilization of the valuable forage resource on winterages, while maintaining biodiversity. To profile the forage quality of winterage, a range of vegetation types have been sampled and analysed to determine their ash, nitrogen, crude protein, oven dry matter, acid detergent fibre, neutral detergent fibre and trace mineral contents. Results show variations in forage quality during the winter grazing season and between vegetation types. Forage quality results suggest that Burren winterages do not meet the nutritional requirements of suckler cows from December to March.

Keywords: burren, grazing, limestone grasslands, forage quality

## Introduction

The Burren is a unique farmed landscape of limestone pavement, grasslands, heaths and wetlands located in the west of Ireland. Approximately 30,000 ha are designated as special areas of conservation (SAC), forming part of Europe's Natura 2000 ecological network of protected sites. Traditional farming practices, in particular winter grazing with minimal external inputs, are important in maintaining the good ecological status of the Burren (Dunford, 2002). Over the past 30 years agriculture in the Burren has undergone substantial change, including the switch from store cattle to suckler beef production and concentration of agricultural activities on agriculturally improved grasslands. The higher nutritional requirements of in-calf cows resulted in many farmers introducing silage onto winter grazed areas (winterage) or housing animals over the winter. Silage feeding leads to animals foraging far less, leading to undergrazing and point source pollution around feeding sites, while the housing of animals results in the abandonment of winter-grazed grasslands and the loss of important management traditions. BurrenLIFE, an EU LIFE-Nature funded project has been set up to develop a new model of sustainable agricultural management for the priority habitats of the Burren. Central to this is the introduction of grazing regimes making use of the valuable forage resource on farms while maintaining biodiversity. The ongoing analysis of the forage quality on Burren winterages is being used to ascertain if Burren winterages meet the nutritional requirements of suckler cows, which are the most prevalent livestock type on the winterages. This aims to encourage sustainable grazing levels on conservation grasslands while meeting the nutritional needs of the grazing animal.

## Materials and methods

Twenty BurrenLIFE monitor farms were selected across the Burren using criteria such as SAC area, types of habitat, grazing levels and farming system. BurrenLIFE monitor farms cover 3097 ha of which 2486 ha are designated as SAC. To profile the forage quality of Burren winterage grasslands, they were divided into 5 broad types based on previous research, which are: (a) *Molinia caerulea* dominant, (b) *Dryas octapetala* dominant (c) *Sesleria caerulea* and *Festuca* sp. dominant, (d) *Calluna vulgaris* approximately 25% cover and (e) *Anthoxanthum odoratum*, *Cynosurus cristatus* and *Dactylis glomerata* dominant. 50 sample areas representing these vegetation types were selected across the monitor farms. Samples were collected every two months during the late summer and winter grazing periods from August to April, from December 2005 until April 2007. Sample locations were located using a GPS and approximately 500 g of forage were cut using clippers. Samples were analysed for ash, N (Kjeldahl nitrogen), crude protein (CP) (N x 6.25), oven dry matter, acid detergent fibre (ADF) and neutral detergent fibre (NDF) (Van Soest analysis) at the Agri-Food and Biosciences Institute, Northern Ireland. Trace mineral analysis was carried out on samples collected in December 2006 using inductively coupled plasma-mass spectroscopy (ICP-MS) to ascertain elemental concentrations (Cu, Mn, Mo, Se, Zn, Ca, K, Mg, P and I) at the Macaulay Institute, Scotland.

The vascular plant species in 2 x 2 m quadrats at each sampling site were recorded and their cover estimated using the Domin scale. These data were used to accurately assign each sampling location to a vegetation type using cluster analysis (PC-ORD vers. 5: relative Euclidean distance measure and wards linkage method) and indicator species analysis.

## Results and discussion

Cluster analysis divided the 50 samples into 6 vegetation types (Table 1). These approximate the M25b (*Molinia caerulea*-*Potentilla erecta* mire, *Anthoxanthum odoratum* sub-community), CG9 (*Sesleria caerulea*-*Galium sternerii* grassland), CG13 (*Dryas octapetala*-*Carex flacca* heath), intermediate between CG9/CG1 (*Sesleria caerulea*-*Festuca* grassland), MG5b (*Cynosurus cristatus*-*Centaurea nigra*, *Galium verum* sub-community) and an enriched MG5 (*Cynosurus cristatus*-*Centaurea nigra* with weedy element) vegetation communities of the national vegetation classification of Britain (Rodwell *et al.*, 1992).

Table 1. Mean  $\pm$  s.e.m. of forage quality variables for Burren winterage vegetation types.

Veg Type (sites)	N	DM g kg <sup>-1</sup>	Ash g kg <sup>-1</sup>	CP g kg <sup>-1</sup>	ADF g kg <sup>-1</sup>	NDF g kg <sup>-1</sup>
			DM	DM	DM	DM
<i>Molinia caerulea</i> mire (5)	37	412.1 $\pm$ 31.8	29.6 $\pm$ 1.8	63.1 $\pm$ 3.8	432.2 $\pm$ 8.5	782.8 $\pm$ 10.4
<i>Sesleria caerulea</i> grassland (7)	56	384.1 $\pm$ 15.9	35.1 $\pm$ 1.5	66.5 $\pm$ 1.7	423.9 $\pm$ 6.2	725.0 $\pm$ 7.8
<i>Dryas octapetala</i> heath (12)	93	383.3 $\pm$ 12.9	38.5 $\pm$ 1.0	72.1 $\pm$ 1.3	427.4 $\pm$ 4.9	711.8 $\pm$ 6.9
<i>Sesleria caerulea</i> - <i>Festuca</i> grassland (10)	74	346.2 $\pm$ 13.7	42.7 $\pm$ 1.3	84.3 $\pm$ 1.8	405.2 $\pm$ 6.4	696.6 $\pm$ 9.9
<i>Cynosurus cristatus</i> - <i>Centaurea nigra</i> grassland (12)	96	303.3 $\pm$ 12.5	58.1 $\pm$ 2.2	95.6 $\pm$ 2.4	390.0 $\pm$ 5.4	698.9 $\pm$ 7.1
enriched <i>Cynosurus cristatus</i> - <i>Centaurea nigra</i> grassland (4)	32	272.6 $\pm$ 23.0	68.1 $\pm$ 5.4	122.8 $\pm$ 6.7	357.0 $\pm$ 9.9	675.4 $\pm$ 14.8
Total	388	349.7 $\pm$ 6.9	45.3 $\pm$ 1.0	82.7 $\pm$ 1.3	408.0 $\pm$ 2.8	711.3 $\pm$ 3.8

Univariate analysis of variance of the forage quality shows that there are significant differences ( $P < 0.01$ ) in the CP, ADF and NDF of the different vegetation types on Burren winterages. The vegetation communities of highest conservation importance and listed as priority habitats for conservation under the EU Habitats directive (i.e. CG 9 and CG13) are dependent on winter grazing to maintain their conservation value (Dunford, 2002). Results in



this study indicate that these grasslands have low average CP and high ADF values on Burren winterages (Table 1). Crude protein values below 70 g kg<sup>-1</sup> DM have been shown to limit fibre digestion in the rumen (Allison, 1985). As expected there is considerable seasonal variation in forage quality (Figure 1). Forage quality decreases as growth slows from August to December as indicated by increases in ADF and decreases in CP values. Forage quality is at its lowest from December to February and improves from February to April. Trace mineral analysis showed that Burren winterage vegetation is deficient in Cu (< 5 mg kg<sup>-1</sup> DM), Se (< 0.10 mg kg<sup>-1</sup> DM), Mg (< 2 g kg<sup>-1</sup> DM) and P (< 2 g kg<sup>-1</sup> DM) (Source: Rogers *et al.*, 2000).

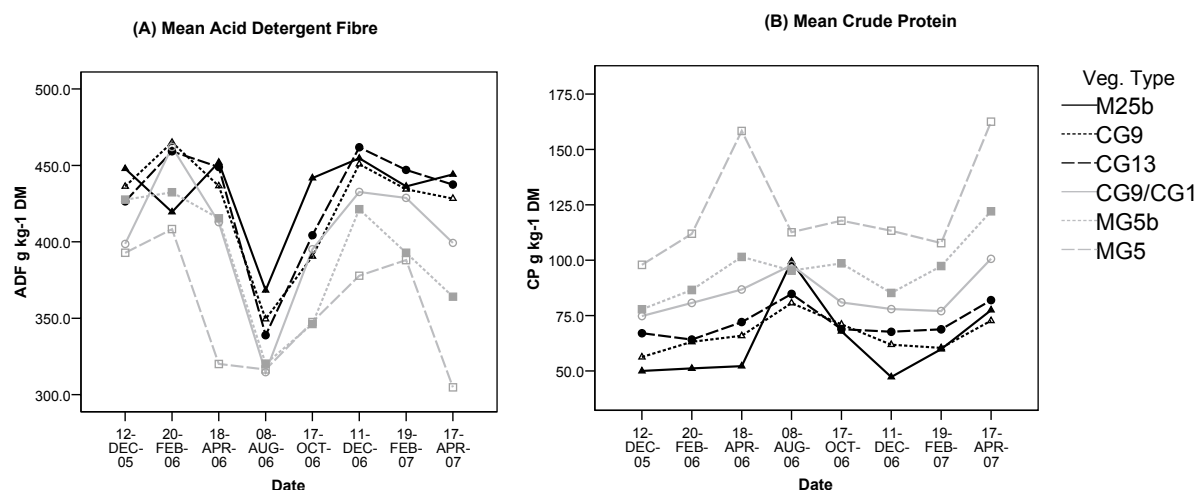


Figure 1: (A) Mean ADF and (B) Mean CP values for Burren winterage types from December 2006 to April 2007.

## Conclusions

The results suggest that Burren winterages do not meet the nutritional requirements of suckler cows, especially from December to March when cows are in late pregnancy. However, this does not take into account selective grazing of more palatable vegetation, which may result in consumption of forage of higher quality than suggested by this study. Supplementation with concentrate feed (replacing existing silage feeding) from December to March could meet the maintenance requirements of cows and increase the supply of crude protein. This can increase the breakdown and rate of passage of poor quality forages through the gut allowing the animal to consume more (Romey and Gill, 2000). As a consequence improving the grazing levels on winterage areas, allowing farmers to graze cows on winterage areas for longer, while meeting their nutritional requirements without resorting to silage feeding or housing. Concentrate supplementation is currently being piloted by the BurrenLIFE project with initial positive feedback from the farmers involved.

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# Management strategies to restore agriculturally affected meadows on peat – biomass and N, P-balances

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## Abstract

In agriculturally affected meadows on peat, soil N and P may be high. To restore such areas and obtain more diverse vegetation the initial management has to include removal of nutrients by a cutting strategy. In 1997 different management strategies were established on an agriculturally affected meadow on peat soil to harvest nutrients and to improve biodiversity. The management options were early and late first cutting with two cuts, combinations of cutting and grazing, addition of K-fertilizer (deep litter or vinasse). The paper presents results on how different strategies influence productivity, nutrient balances and botanical composition on a dry matter (DM) basis. Addition of potassium increased productivity and by cutting it was possible to remove substantial amounts of nutrients. Without any fertilizer the cutting strategies also increased the nutrient removal and affected botanical composition. In 2006-07 the percentage of cultivated grass species in the harvested DM was lower under late cutting than early first cutting with two cuts.

Keywords: restoration, peat soil, deep litter, vinasse, botanical composition

## Introduction

In agriculturally affected meadows on peat, soil N and P may be high. To restore such areas and obtain more diverse vegetation the initial management has to include removal of nutrients by a cutting strategy. Under continuous summer grazing 8 kg N, 2 kg P and 1 kg K ha<sup>-1</sup> were removed in a Danish meadow (Hald *et al.*, 2003). By adding the limiting nutrient, here K, it seems possible to collect more N and P by a cutting strategy. It was found that the addition of deep litter and two cuts a year resulted in a lower level of plant available N in spring than continuous grazing (Nielsen and Hoffmann, 2005). Deep litter adds surplus K, and it is more efficient to use vinasse (which contains 21% K, 0.5% N and 0% P). Vinasse is also allowed in organic farming systems in Denmark. The aim was to examine the effect on dry matter (DM) yield, nutrient balances and botanical composition by applying different management strategies, as well as through the effect of changing the source of K from deep litter to vinasse.

## Materials and methods

The investigation was carried out on long term grassland from 1997 to 2007. Before 1990 the area had been used as intensive grassland, and from 1990 to 1997 the management was extensive cutting and grazing without fertilizer. The management experiment was laid out in 1997 in three blocks with a range of treatments: late (Treatment b) and early (Treatment g) cutting with two cuts; grazing following early cut (Treatment e) and late cut (Treatment f), and addition of K-fertilizer (deep litter or vinasse) (Treatment h) at a low diversity area. Also, grazing only and abandoned plots were established. The size of plot was 144 m<sup>2</sup> for cutting only, and the plots with grazing were laid out in connection with bigger paddocks. The

following changes were made to Treatment h: deep litter and late cut in 1997-2004, vinasse and early cut in 2005-2007.

Herbage yield and nutrient content were measured in 1997-2003 and 2006-2007. Percentages of single species as well as the fraction of pooled dead material were calculated on DM-basis. The upper 100 cm soil had 65% organic matter and 2.8% total N. In 1997-2000 the water level was measured monthly and varied from -49 to +13 through the year (Hald *et al.*, 2003).

## Results and discussion

With one cut only, the highest DM yield and greatest removal of nutrients was obtained by late first cutting (Treatment f), significantly different from early cutting (Treatment e), in 1997-2003 (Table 1). With two cuts the highest yield and removal of nutrients was obtained with vinasse in 2006-2007, where 180 kg N and 20 kg P ha<sup>-1</sup> were removed by cutting.

Table 1. DM-yield, N- and P-balance in 1997-2003 and in 2006-2007. Data for cutting only. Different letters indicate significant difference ( $P < 0.05$ ) within years of comparison. (Cut E: first cut on 10 June; Cut L: first cut on 15 July).

	Average of first 7 years of management (1997-2003) – deep litter						Average of 10th and 11th year of management (2006-07) – vinasse					
	Cut E Graz. e	Cut L Graz. f	Cut E Cut g	Cut L Cut b	Cut L <sup>1</sup> Cut h	Lsd <sup>2</sup>	Cut E Graz. e	Cut L Graz. f	Cut E Cut g	Cut L Cut b	Cut L <sup>1</sup> Cut h	Lsd <sup>2</sup>
<b>DM</b> t ha <sup>-1</sup> y <sup>-1</sup>	b 2.7	a 4.0	b 4.3	b 4.1	a 7.1	1.0 1.1	2.2	3.0	b 4.5	b 6.2	a 9.4	2.0 2.5
<b>N-balance</b> kg N ha <sup>-1</sup> y <sup>-1</sup>	a -59	b -77	b -102	ab -97	a -65	5 21	-47	-51	a -98	a -132	b -180	27 44
<b>P-balance</b> kg P ha <sup>-1</sup> y <sup>-1</sup>	a -8	b -11	b -13	b -12	a -1	1 3	-5	-7	a -11	ab -15	b -20	5 6

<sup>1</sup>Deep litter and late cut in 1997-2004, vinasse and early cut in 2005-2007; in 2004-2005 management only and no analyses.

<sup>2</sup>Lsd: Least significant difference.

The grass species have been grouped as cultivated grasses (*Dactylis glomerata* L., *Festuca rubra* L., *Lolium perenne* L., *Phleum pratense* L., *Poa pratensis* L. and *Poa trivialis* L.) which are used to reseed swards, and as natural grasses. Rush is here predominantly *Juncus effusus* L. and ‘other herbs’ are predominantly *Ranunculus repens* L. Mosses were also found but most often at < 1% of the total material. The botanical composition at first cut is shown in Table 2. The cultivated grasses decreased in general, and in 2006-2007 their contribution was lower in Treatments b and f than in Treatment g. *Poa pratensis* managed to retain its contribution better in Treatment g than in the other treatments. *Poa trivialis* managed better in Treatment h than in b and f. The group of natural grasses managed better in h than in g, mainly because of *Phalaris arundinacea* (L.)R.Br. The contribution of *Holcus lanatus* L. was very low in 1997, but it increased to 12-14% in 2006-2007 in Treatments b, f and h. ‘Other herbs’ increased more in h than in b. As expected the percentage of dead material was higher in the strategies with a late, compared with an early, first cut.

K addition affected the botanical composition. The increase in *P. arundinacea* resulted in a tall and dominating vegetation, and this is not a desirable management for swards that have a

high botanical nature quality. Where the botanical nature quality is low and the level of N and P is high, it seems relevant to use potassium to collect surplus of other nutrients by cutting over a span of years. If the nutrient status of the soil decreases to a level where the vegetation becomes scarcer, it may be possible to introduce more species to the sward, e.g. by grazing the area together with a high botanical nature quality sward. How long the management with addition of K and cutting is necessary depends on the soil nutrient status and whether the specific area is affected from the adjacent agricultural areas.

Table 2. Botanical composition on a percent-DM basis at first cut. Different letters indicate significant difference ( $P < 0.05$ ) within species or species group in 1997 or 2006-2007. (Cut E: first cut on 10 June; Cut L: first cut on 15 July).

1 <sup>st</sup> Treatm. 2 <sup>nd</sup> Treatm.	First year of management (1997)						Average of 10th and 11th year of management (2006-2007)					
	Cut L	Cut E	Cut L	Cut E	Fertil Cut L <sup>1</sup>	Lsd <sup>2</sup>	Cut L	Cut E	Cut L	Cut E	Fertil Cut E <sup>1</sup>	Lsd <sup>2</sup>
	b	e	f	g	h		b	e	f	g	h	
<b>Cult. Grass</b>	37	66	35	57	41	34	11	19	9	31	20	16
<i>P. pratensis</i>	31	52	29	50	34	36	5	8	3	21	5	10
<i>P. trivialis</i>	3	5	2	3	2	6	3	10	2	8	13	9
<b>Nat. grass</b>	35	28	31	31	26	35	58	62	61	51	69	17
<i>P. arundinacea</i>	20	19	20	17	15	25	8	9	4	17	41	11
<i>D. caespitose</i>	< 1	< 1	< 1	7	< 1	10	28	36	38	8	1	14
<i>H. lanatus</i>	< 1	< 1	< 1	1.6	< 1	2	12	2	14	1	13	9
<i>A. stolonifera</i>	5	5	< 1	3	< 1	8	7	2	2	8	3	10
<i>G. fluitans</i>	3	< 1	< 1	< 1	< 1	3	4	6	3	7	2	9
<i>A. geniculatus</i>	5	2	8	2	6	10	< 1	7	< 1	10	< 1	9
<b>Rush<sup>3</sup></b>	< 1	1	< 1	< 1	< 1	2	3	5	2	4	1	5
<b>Other herbs<sup>4</sup></b>	2	< 1	< 1	< 1	< 1	2	3	4	3	3	6	3
<b>Dead mat.</b>	26	5	34	12	32	14	24	10	25	9	4	8

<sup>1</sup>Deep litter and late cut in 1997-2004, vinasse and early cut in 2005-2007. <sup>2</sup>Lsd: least significant difference.

<sup>3</sup>Mainly *J. effusus*. 4) Mainly *R. repens*.

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# ***Betula pendula*, *Pinus sylvestris* and *Quercus robur* trees in Swedish semi-natural pastures – effects on plant diversity**

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## **Abstract**

The positive versus negative effects on plant species diversity by the presence of trees are often discussed when considering restorations in Swedish semi-natural grasslands. Therefore, plant diversity was studied under trees of *Betula pendula*, *Pinus sylvestris* and *Quercus robur*. For each tree species 30 individuals were studied and four plots were placed in a line from the trunk into the open vegetation. In each plot (20 x 20 cm) vegetation height, litter depth, and abundance of all species and seedlings was noted, as well as the number of flowering herbs and grasses. The number of species was higher in the open vegetation than under the tree canopies for all tree species. Forest species and shade tolerant species like *Deschampsia flexuosa* and *Anemone hepatica* increased under the tree crowns. Species with high nitrogen status were most common under *Q. robur* trees. The vegetation was denser in the open areas than under tree canopies. Litter covered 50-60% of the plots under the tree crowns and about 15% in the open vegetation. The number of flowering herbs and seedlings increased with the distance from the trunk. In conclusion, trees were positive for forest species and shade tolerant species and increase the heterogeneity in semi-natural grasslands.

Keywords: Trees, semi-natural pasture, grazing, plant reproduction

## **Introduction**

Encroachment of woody species in semi-natural grasslands following abandonment is a general phenomenon in the temperate zone and overgrowth of trees and shrubs is a threat to many grassland species (Dzwonko and Loster, 2007; Pärtel and Helm, 2007). Under dense tree canopies the number of light-demanding species decreases and they are replaced by shade-tolerant forest species (Curt *et al.*, 2003). Trees can also reduce radiation, plant cover and plant biomass under their crowns (Facelli and Pickett, 1991). Moreover, trees influence the soil characters beneath their canopies; they can alter soil pH, increase soil nutrients and lower soil temperature (Dahlgren *et al.*, 1997). In Sweden clearing of trees and shrubs in semi-natural grasslands is prescribed to enhance biodiversity. Restoration activities such as clearing and reintroduced grazing are successful in increasing plant species richness (Willems, 2001; Rosen and Bakker, 2005). However, some studies have found that the presence of trees and shrubs in semi-natural grasslands increases species richness of plants (Söderström *et al.*, 2001; Lindborg and Eriksson, 2004). Although effects of both tree encroachment and restoration activities have been studied, the effects of free-standing trees on the grassland flora have seldom been studied. How do plant species richness, plant reproduction and the vegetation cover vary along a gradient from below the tree canopy to adjacent open areas? Can species patterns be attributed to functional traits such as Ellenberg values for light and nitrogen status? We hypothesize that trees reduce the vegetation cover, the number of light demanding species and plant reproduction.

## **Materials and methods**

Twenty semi-natural pastures in south central Sweden (59°20'–60°22'N; 16°09'–18°16'E)

were used as study sites. The pastures were on average 13 ha in size and grazed by beef or dairy cattle. The vegetation was unfertilized and uncultivated and had high conservation values (Persson, 2005). The vegetation was studied under 90 free standing trees of *Betula pendula*, *Pinus sylvestris* and *Quercus robur*, 30 individuals of each tree species. Only trees with a minimum of 60 cm in perimeter were chosen. The distance between the trunk and the crown edge was measured and four plots (20 x 20 cm) were placed along a line from the trunk into the open vegetation. The first plot was placed next to the trunk and the second plot was placed at 1/3 of the distance from the trunk to the crown edge and the third plot at 2/3 from the trunk. The fourth plot was placed 2 m outside the crown edge. At each tree two lines were studied, one was facing north and one facing south. In total 720 plots were studied, i.e. 240 plots per tree species. In all plots vegetation height and litter depth were measured. The cover of vegetation, bare ground and/or litter in the plots was estimated on a percentage basis. Litter consisted generally of leaf or needle litter, bark or dead vegetation. The presence of all species in each plot was noted and the number of flowering herbs, flowering grasses and seedlings was counted. For most herbs single buds, flowers or fruits were counted and for herbs with panicles, cymes, composite umbels or racemes, these were counted. For grasses, panicles were counted. Seedlings were not determined to species. Plant species patterns were explained by using Ellenberg values for light and nitrogen (Ellenberg, 1991).

## Results and discussion

The total number of species was significantly lower beneath the tree canopies for all three species, *B. pendula*, *P. sylvestris* and *Q. robur*, than in adjacent open areas (ANOVA,  $P < 0.001$  in all cases). Light-demanding species like *Achillea millefolium*, *Agrostis capillaris* and *Trifolium repens* increased with increasing distance from the tree trunks, while shade-demanding species like *Deschampsia flexuosa* and *Anemone hepatica* were most common under the tree canopies (Table 1). Species like *Alopecurus pratensis*, *Anthriscus sylvestris* and *Taraxacum vulgare*, that indicate sites with high nitrogen status were most common under *Q. robur* trees, and species like *Festuca ovina*, *Galium boreale* and *Vaccinium vitis-idaea*, with low Ellenberg values for nitrogen, were mainly found under *B. pendula* and *P. sylvestris* trees (Table 2). The number of reproducing herbs was higher in open areas than below the tree canopies for all tree species ( $P < 0.01$  in all cases). No differences in number of reproducing grasses between open pasture and beneath tree crowns were found for any of the tree species ( $P > 0.05$ ). The number of seedlings was lower under *B. pendula* and *Q. robur* canopies than in adjacent open areas ( $P < 0.001$ ) and the same tendency was found for *P. sylvestris* ( $P = 0.058$ ). There was no difference in vegetation height or cover of bare ground beneath the tree crowns compared to the area outside the trees for any of the tree species ( $P > 0.05$ ). The vegetation covered 83% in open pasture and 40-70% of the area under the tree canopies ( $P < 0.001$ ). Litter covered about 15% in the open vegetation and between 30% and 60% beneath the tree crowns and the differences were significant for all tree species ( $P < 0.001$ ). The litter layer was thicker beneath *B. pendula* and *P. sylvestris* crowns than in the open areas ( $P < 0.001$ ), but the difference was not significant in the case of *Q. robur* trees ( $P = 0.589$ ).

Table 1. Mean abundance of five species compared at four distances from the tree trunk, and Ellenberg values for light for each species.

Distance from tree trunk	0	1/3	2/3	Open	Ellenberg light
<i>Achillea millefolium</i>	4.2	10.5	16.0	20.3	8
<i>Agrostis capillaris</i>	6.8	13.7	18.8	24.7	7
<i>Trifolium repens</i>	2.0	5.0	9.3	15.0	8
<i>Anemone hepatica</i>	0.3	1.0	1.0	0.3	4
<i>Deschampsia flexuosa</i>	7.2	3.2	2.5	2.8	6

Table 2. Mean abundance of six species compared for the canopies of the three studied tree species (*Q. robur*, *B. pendula* and *P. sylvestris*) and their Ellenberg values for nitrogen.

	<i>Quercus robur</i>	<i>Betula pendula</i>	<i>Pinus sylvestris</i>	Ellenberg nitrogen
<i>Alopecurus pratensis</i>	3.6	0.3	0.3	7
<i>Anthriscus sylvestris</i>	6.5	0.6	1.3	8
<i>Taraxacum vulgare</i>	5.3	2.3	1.6	8
<i>Festuca ovina</i>	2.1	5.9	5.3	1
<i>Galium boreale</i>	3.6	3.8	8.5	1
<i>Vaccinium vitis-idaea</i>	0.0	2.8	0.5	1

## Discussion

All tree species reduced the number of plant species beneath their canopies and our results correspond with previous studies that trees are negative for light-demanding grassland plant species and positive for shade-demanding and forest species. Species that indicate nitrogen-rich sites were more common under *Q. robur* trees than under *B. pendula* and *P. sylvestris* trees and this may be due to biochemical processes by which oaks concentrate nutrients beneath their canopies (Dahlgren, 1997). Presence of trees in grasslands increases the heterogeneity in shade conditions and nitrogen status, and thereby trees probably contribute to increase the total plant-species richness. When restoring overgrown pastures it is important to consider shade and nutrient gradients, and to create open areas, areas with free-standing trees and areas with denser tree stands. Pollarding of the deciduous trees *B. pendula* and *Q. robur* trees may also be used. This will increase light conditions for grassland plants and, at the same time, create suitable substrates for many red-listed species like lichens, bryophytes, insects and fungi (Gärdenfors, 2005).

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# Management strategies to reduce regrowth species and increase biodiversity in semi-natural grasslands in central Norway

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## Abstract

In central Norway, *Anthriscus sylvestris*, *Deschampsia cespitosa* and *Alnus incana* are well known as regrowth species that colonize abandoned areas. They often dominate abandoned areas and reduce the species biodiversity. The objectives of the field studies described were to find appropriate management strategies for reducing the plant cover of *A. sylvestris*, *D. cespitosa* and *A. incana* in semi-natural grasslands, and thereby to conserve or increase the biodiversity. In road verges dominated by *A. sylvestris*, management studies by different mowing strategies were recorded during four years. In abandoned grasslands the responses of *D. cespitosa*, *A. sylvestris* and *A. incana* to four different grazing regimes were investigated over four years. Results from these studies show that areas dominated by these species require specific, extraordinary, intensive and long-term management strategies.

Keywords: biodiversity, *Alnus incana*, *Anthriscus sylvestris*, *Deschampsia cespitosa*

## Introduction

In central Norway, *Anthriscus sylvestris*, *Deschampsia cespitosa* and *Alnus incana* are well known as regrowth species that colonize abandoned areas. They often dominate abandoned areas and thereby reduce biodiversity (Losvik, 1988). They are difficult to control when restoring (by cutting, mowing and/or grazing) abandoned semi-natural grasslands. Restoration might be necessary to control these species and re-establish and maintain some types of semi-natural grasslands (Hobbs and Harris, 2001). The objectives of these field studies were to find appropriate management strategies to reduce the plant cover of *A. sylvestris*, *D. cespitosa* and *A. incana* in semi-natural grasslands and thereby conserve or increase the biodiversity.

## Methods

Different field studies were performed for each species. All the studies were made in Trøndelag Counties in central Norway. On road verges dominated by *A. sylvestris* management studies by different mowing strategies were recorded during four years on the same 45 1 m<sup>2</sup> quadrats each year. Treatments were: mowing early (x 2) and mowing late (x 2) followed by removing/not removing the biomass, and a control; percentage plant cover was measured in each. In abandoned grasslands the response of *D. cespitosa*, *A. sylvestris* and *A. incana* to four different grazing regimes (continuous and rotational grazing by sheep and continuous and rotational grazing by cattle), and a control, was investigated for four years on the same 120 1 m<sup>2</sup> quadrats each year. Plant cover was measured by frequency. All the plant cover analyses were performed using *The SAS<sup>®</sup> system for Windows<sup>™</sup>* release 8e. To investigate trends over time, a mixed model procedure for repeated measures of plant cover was modelled as a polynomial function of time (Littell *et al.*, 1996). This was done for each of the treatments, i.e. all the combinations of mowing and grazing regimes. For each site, contrasts were used to investigate which treatments differed significantly. Because the variable time is quantitative, cover can be modelled as a polynomial function of time. Because (with exception of the response of *A. sylvestris* to the different grazing regimes), both plots and



statistical tests made of the data indicated that there was a quadratic correlation between cover and time, quadratic regression models were fit to the data (Littell *et al.*, 1996). This was done for each treatment, i.e. all combinations of mowing and grazing regimes. For each site, contrasts were used to investigate which treatments differed significantly. In the same way, a linear regression model was fit to the data of response of *A. sylvestris* to the grazing regimes.

## Results and discussion

*Anthriscus sylvestris* was increased by mowing twice per season (independent of biomass removal) (Figure 1a) and also in sheep-grazed areas (Figure 1b). Both mowing and sheep grazing removed flowering stems before seed dispersal. To compensate for the loss of seed dispersal, vegetative regeneration by lateral shoots increased (Rosef and Bele, 2007).

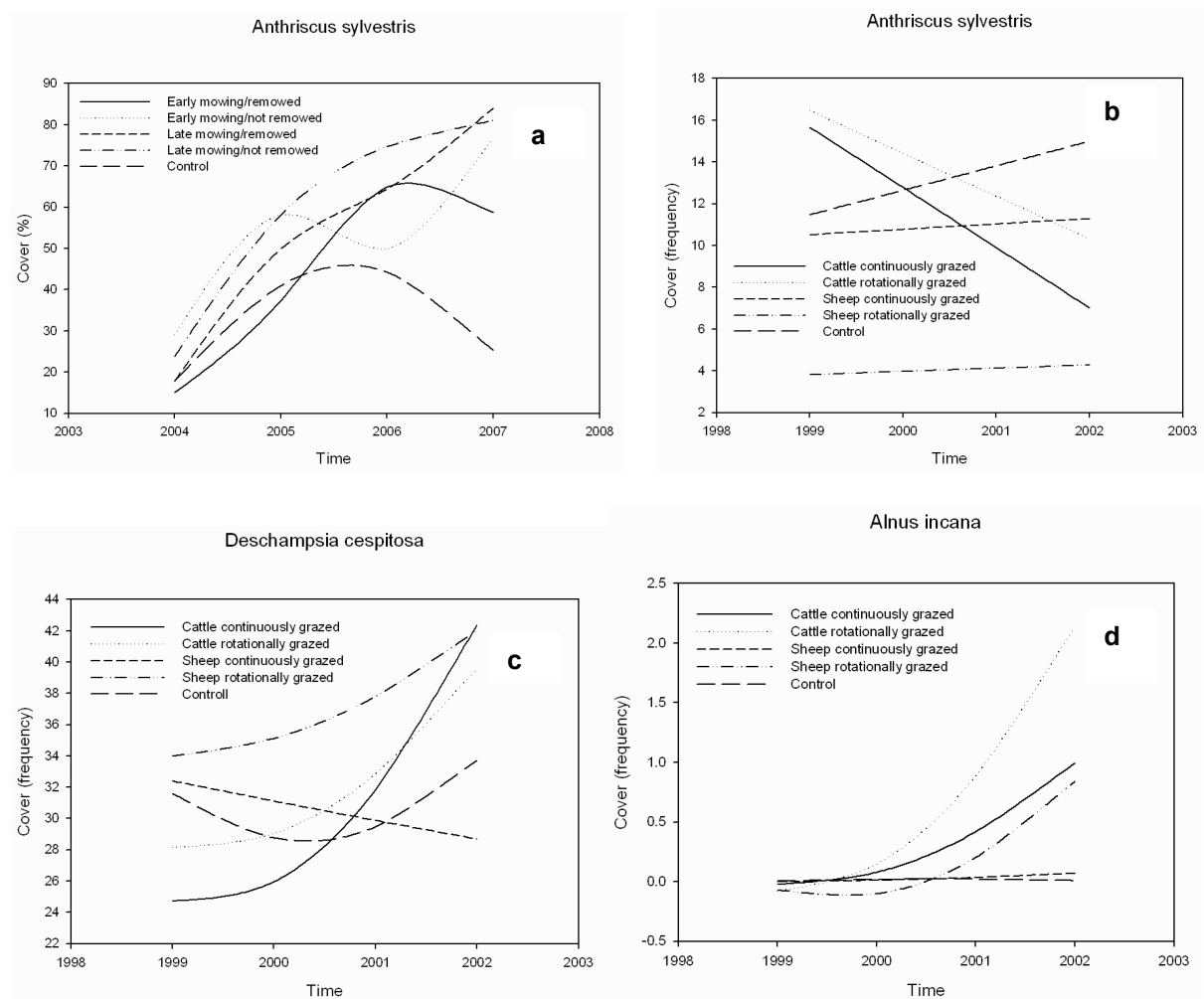


Figure 1 a-d. Plant cover of *A. sylvestris* with the four different mowing treatments and the unmown control (a), and plant cover of *A. sylvestris*, *D. cespitosa* and *A. incana* for the four grazing treatments and the ungrazed control (b-d) all results during four years.

In continuous cattle-grazed areas the cover of *A. sylvestris* decreased, probably due to trampling effects and damage to the root system (Grime *et al.*, 1988). Because *A. sylvestris* is a nutrient demanding species, fertilization must be avoided. The biomass must be removed after mowing, both to decrease the nutrient levels and because seedling establishment of this species has a positive response to the litter layer (Hovstad and Ohlsson, 2007). The results did not show any effect of removing biomass, probably because the study period was only four years and fertilizer from surrounding fields might have been added. Management strategies to

decrease plant cover of *A. sylvestris* must take into account that the two different regeneration strategies (lateral shoots and seeds) varies with life-cycle/age and management activities (Johansson and Hedin, 1995) and it must be planned for a long period of time (more than four years). The management regime should be intensive during the whole season (Parr and Way, 1988). Grazing by cattle and rotational grazing by sheep increases the cover of *D. cespitosa* and *A. incana*, while continuous grazing by sheep did not have any effect (Figures 1c and d). The response of *D. cespitosa* and *A. incana* to grazing should be considered as at least two different processes. Grazing both by cattle and sheep may to a certain extent reduce the cover of *D. cespitosa*, while grazing is usually not enough to control the growth of *A. incana*. The effect of trampling may benefit the distribution as well as the cover of *D. cespitosa* and *A. incana*, improving conditions for germination, even though trampling generally is expected to damage the vegetation. It is important to keep this in mind when restoring abandoned grasslands where *D. cespitosa* and/or *A. incana* are dominant species. If the target is to control the species, successful management will depend on an adequate grazing pressure and the total cover at the beginning of the restoration. At the same time it is important to avoid creating bare ground, i.e. minimizing damage by trampling (Rosef, 2004; Rosef *et al.*, 2007). Continuous hard grazing by sheep may decrease the cover of *D. cespitosa* and *A. incana*; however, excessively hard grazing pressure may be inadvisable for animal welfare reasons.

## Conclusion

Field studies of *A. sylvestris*, *D. cespitosa* and *A. incana* in central Norway showed that these species are well adapted to compensate for management activities intended to reduce the plant cover in a restoration process. Consequently, they need species-specific, extraordinary, intensive and long-term management strategies. This must be considered in the planning of management activities in abandoned areas dominated by these species.

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# Analysis of the influence of some invasive plants species on the pastoral value of western Romanian grasslands

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## Abstract

Pastoral value represents the forage potential of grassland, this being determined by the specific quality index of the species that are forming the vegetation cover. The purpose of this work is to analyse the influence of invasive plant species on grassland pastoral value. This study was carried out during 2003-2005 on 26 grasslands in western Romania. The analyses were made of 15 plant species considered on the basis of previous research to be invasive species in our plots. Pastoral value of the vegetation analysed here was found to be decreasing in successive years of the study, and the surface covered with our target species was increasing. Most of the analysed species analysed in this work are determining the decrease of the pastoral value of the grassland.

Keywords: invasive species, pastoral value, correlation, grassland

## Introduction

The grasslands studied in this research show different pastoral values starting with low to average. The most frequent invasive plant species in western Romanian permanent grasslands belong to the families: *Rosaceae* (*Prunus spinosa*, *Crataegus monogyna*, *Rosa canina*, *Rubus caesius*), *Fabaceae* (*Sarothamnus scoparius*, *Amorpha fruticosa*), *Juncaceae* (*Juncus effusus*), *Asteraceae* (*Carduus acanthoides*, *Carlina vulgaris*, *Carthamus lanatus*, *Cirsium undulatum*), *Apiaceae* (*Eryngium campestre*), *Dipsacaceae* (*Dipsacus fullonum*), *Euphorbiaceae* (*Euphorbia cyparissias*), and *Dennstaedtiaceae* (*Pteridium aquilinum*) etc. The role of ecosystem disturbances on the promotion of invasive plant species is essential. Undergrazing, overgrazing and the lack of the minimal maintenance works on grasslands leads to the proliferation of invasive plant species. Environmental changes are also determined by the change of the pressure of some anthropogenic factors (use, fertilization, maintenance works, management), which are influencing the botanical composition of the permanent grasslands.

The actual state of most Romanian grassland is greatly influenced by the agricultural activities from the communist period, and from the recent period also, because after 1989 many of these surfaces were inappropriately used or abandoned. Also, the number of animals that are using these permanent grasslands has decreased greatly. These conditions have determined major changes in the vegetation structure of the Romanian grasslands. Thus, favourable conditions have occurred for the spreading of some species with invasive traits that have negative influence on the initial valuable vegetation, some of them becoming dominant in the vegetation cover (Sărățeanu and Moisuc, 2004). The main objective of this research is to obtain evidence of the influence of invasive plants species on the pastoral value under the ecological conditions of western Romania.

## Material and methods

This study was carried out in 2003-2005 on 26 permanent grasslands situated in places with different environmental conditions from Banat region (western Romania). The data were collected once a year. The surface of the grasslands studied has different degrees of cover of invasive species, and different vegetation features (dominance, biodiversity etc.) and different

pastoral values. Research plots were placed at altitudes between 87 and 370 m, on soils with pH between 5.4 and 8.0 pH. The mapping of the aerial projection of plants on 100 m<sup>2</sup> (10 m x 10 m) was done by dividing the studied surface into 100 sub-plots (1 m x 1 m). For each sub-plot we have evaluated the area covered with invasive plants (m<sup>2</sup>). The data obtained in this way helped us to analyse the spatial distribution, and to calculate the coverage index for the studied species, which is the ratio of the total area covered by the target species (sum of the areas covered in 100 sub-plots) to the total area of the plot (100 m<sup>2</sup>). The pastoral value is calculated considering the specific indexes and has values between 0 and 100, when 0 represents grassland without any forage value, and 100 represents ideal grassland. Vegetation biodiversity is calculated with Shannon-Wiener index, and the botanical composition is determined with square meter method. The statistical methods used were correlation analyses.

## Results and discussion

The grassland analysed in this research has different pastoral values as is shown in Figure 1. The graph shows that the pastoral value of the grassland is decreasing from one year to another, their vegetation being implicated in degradation process.

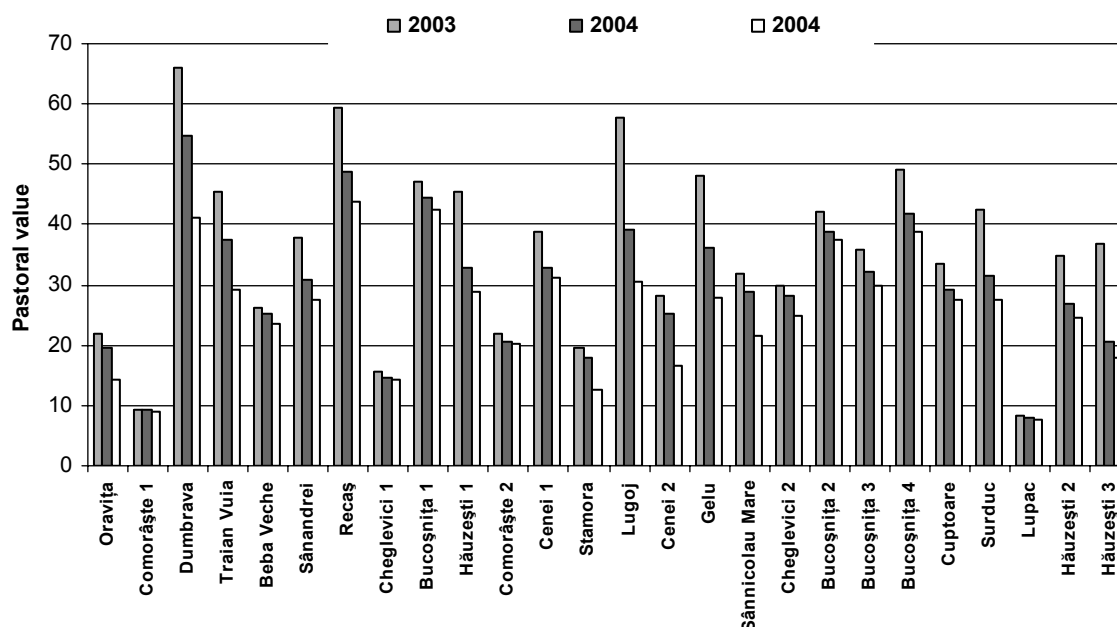


Figure 1. Pastoral value of grasslands (2003-2005).

When we are referring to the pastoral value we consider the vegetation biodiversity too, because the structure of the vegetation cover (respectively the presence of some certain species) is influencing the pastoral value. In Table 1 are represented some correlations between the coverage index of the invasive species and the pastoral value, and Shannon-Wiener index of the grassland. The greatest correlation coefficients obtained for pastoral value are obtained for *Eryngium campestre*, *Dipsacus laciniatus*, *Carlina vulgaris*, *Xanthium spinosum*, *Carthamus lanatus* and *Amorpha fruticosa*. In the case of 10 species from 15, the correlation is negative and conforms to the *r* values obtained between the coverage index and pastoral value, this fact supporting the hypothesis that the increase of the coverage index of invasive species is determining the decrease of the pastoral value. Another aspect analysed in Table 1 is the correlation between the coverage index of the invasive species and the Shannon-Wiener index. There is also a positive interdependence in the case of eight species; they are determining the increase of the biodiversity index. This increase is determined in

reality by a greater number of annual weed species. There is a great interdependence among these two variables conform to the obtained results. Other researchers (Wiser *et al.*, 1998; Stohlgren *et al.*, 1999; Smith and Knapp, 2001) have obtained positive correlations between biodiversity and plants species invasiveness. Moisuc *et al.* (2000, 2001) obtained negative correlation coefficients between the pastoral value and Shannon-Wiener index, and explained that the great incidence of the annual weeds is providing increased biodiversity indexes.

Table 1. The correlations between the coverage index of the invasive species and the pastoral value of the grasslands and the biodiversity index (Shannon-Wiener index).

No.	Species	<i>r</i> (coverage index and pastoral value)	<i>r</i> (coverage index and biodiversity index)
1	<i>Euphorbia cyparissias</i> L.	-0.58 <sup>0</sup>	0.03
2	<i>Juncus effusus</i> L.	-0.64 <sup>0</sup>	-0.05
3	<i>Carduus acanthoides</i> L.	-0.24	0.92 ***
4	<i>Eryngium campestre</i> L.	-0.85 <sup>000</sup>	0.55 *
5	<i>Dipsacus laciniatus</i> L.	-0.89 <sup>000</sup>	-0.89 <sup>000</sup>
6	<i>Carlina vulgaris</i> L.	-0.85 <sup>000</sup>	0.94 ***
7	<i>Xanthium spinosum</i> L.	-0.95 <sup>000</sup>	0.89 ***
8	<i>Carthamus lanatus</i> L.	-0.88 <sup>000</sup>	0.87 ***
9	<i>Pteridium aquilinum</i> (L.) Kuhn	-0.14	-0.51 <sup>0</sup>
10	<i>Rosa canina</i> L.	-0.79 <sup>00</sup>	0.71 **
11	<i>Crataegus monogyna</i> Jacq.	-0.29	-0.02
12	<i>Prunus spinosa</i> L.	-0.73 <sup>00</sup>	0.78 **
13	<i>Rubus caesius</i> L.	-0.02	0.87 ***
14	<i>Amorpha fruticosa</i> L.	-0.89 <sup>000</sup>	-0.53 <sup>0</sup>
15	<i>Sarothamnus scoparius</i> (L.) Wimmer	-0.35	-0.46

## Conclusions

The increase of the coverage index of most of the invasive species of grassland is determining the decrease of its pastoral value, and the increase of the biodiversity index. The increase in plant species biodiversity is due to the increase of the number of the annual weed species.

## Acknowledgements

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# **Biodiversity: the process from scientific results into farming practice**

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## **Abstract**

How to implement scientific knowledge into farming practice? How to develop an incentive system that convinces farmers to 'produce' ecology? In Switzerland a participatory process provides sustainable solutions; it is presented here using the example of a recent successful project on pasture management.

AGRIDEA has been mandated by the Swiss Federal Office for Agriculture to develop from scientific studies practicable and easily understandable criteria that are representative of the high ecological value of pastures. During visits and by holding workshops with farmers, advisers, government officials and controllers, many discussions were held to improve the mutual understanding for the claims of biodiversity and the limitations of agricultural realities in managing pastures. The result is a governmental ordinance which pays subsidies for pastures with a high diversity of flora and landscape features such as special bushes, trees or rocky areas. Furthermore, agricultural circles have been made aware of the need for improved ecological quality through adaptations in grazing and management practices.

Keywords: biodiversity, ecological quality, pastures, grazing management, transdisciplinarity, participatory process

## **Introduction**

Since 1993 Swiss farmers are obliged to manage 7% of their agricultural land as ecological compensation areas to get direct payments by the state. If they choose suitable sites with a high ecological value they are rewarded by additional subsidies, legalized by a new law, the Ordinance on Ecological Quality (ÖQV). This ordinance had been applied on meadows, orchards and hedges that corresponded to defined criteria.

In a further step the Swiss Federal Office for Agriculture intended to apply the ÖQV also on pastures with high ecological diversity. The ecological value of nutrient-poor grazing land had been recognized since several years (Schmid *et al.*, 2001), but there was no scientific basis for defining suitable criteria. A scientific mandate carried out by the Agroscope Reckenholz-Tänikon in collaboration with AGRIDEA on 180 pastures led to a list of indicator plants and animal species that were correlated with a high ecological quality. Furthermore, information was about landscape features and pasture management (Walter *et al.*, 2007).

The question was how to implement scientific results into a programme usable for assessment and inspection, valid for all regions and altitudes of Switzerland and applicable by trained farmers. This development process was carried out by AGRIDEA.

The aim and the challenge were to find criteria accepted by agricultural stakeholders and a reproducible method to judge the ecological value of a pasture. It had to be sure that the definitive method honoured the 'right' pastures with a high biodiversity and at the same time with a sustainable management. How to enhance biodiversity by encouraging the own responsibility of the farmers?

## Methodology

Based on the scientific results a first draft of plausible criteria, evaluation keys and methods was checked and improved on by various experts of flora and fauna. The resulting methodology was adapted and set out in an iterative process by:

- checking the biological and practicable aspects of the method in about 20 pastures in different regions of Switzerland
- testing the method with representatives of agricultural extension services, inspection organizations, government officials and stakeholders of nature conservation
- consulting biological and agricultural experts in several steps

This continuous and participatory process involving demands of agricultural extension, land use management and inspection should enable the development of a methodology that could be widely accepted and realizable and remain compatible with the claims of biodiversity.

## Results and discussion

Agricultural stakeholders pleaded for considering features such as bushes, trees, rocks, stones, streams etc. as a part of the assessment method. They had understanding towards the criteria used for fauna controls but requested for reasons of practicability, that indicative faunal groups or species could be evaluated independent of growing season and weather conditions in one single inspection. Regarding these presuppositions it was decided to abandon fauna as a directly controlled criterion. Frequency of unfavourable weather conditions such as annual fluctuations of faunal population could not ensure reliable results.

As a compensation, the character of features had to represent the claims of most characteristic animals living in pastures. Different experts defined the ecological requirements of animal groups such as birds, lizards, different insect groups. This was taken into consideration by developing a key of landscape features in the form of a points system. Thorny bushes and trees that were especially preferred during the breeding stages were favoured. Also running water, small walls, piles of stones and periodically ungrazed patches received points. A further aspect stressed the distribution of features: grazing habitat with homogeneously distributed structures represent a sustainable management system of pastures and were valued higher than sites with large and dense patches of bushes and trees.

To the representatives of agricultural practice, sustainable management of pastures was a dominant issue. Therefore they wanted to limit the area of problematic species like brambles and bracken strictly and to tolerate only a small covering of bushes.

Some general conditions were proposed – beyond the proof of flora and landscape features – in order to release subsidies.

- The use of artificial fertilizers and manure in addition to the grazing animals should be prohibited.

- Scientific results had shown that the ecological value of pastures was inversely correlated to livestock density and to the duration of grazing. Long intervals between grazing periods were important, especially to fauna biodiversity. Nevertheless, it did not seem reasonable to regulate pasture management uniformly as each pasture is individual in growth, availability of feed, livestock numbers, grazing dates and other parameters. It is the responsibility of the farmers to maintain the ecological value by observing themselves the impact of management on the flora and fauna.

- Evaluation of project data confirmed also that grazing by cattle leads, on average, to a higher biodiversity than that by grazing sheep. However, it was not indicated to exclude certain types of grazing animals when legitimizing subsidies.

- In all ecological compensation areas being paid subsidy, the use of herbicides is prohibited. This of course means physical effort to manage the tree and bush population in order to sustain and guarantee habitat. The challenge to the farmer is to save the ecologically valuable species and to reduce only those plants which tend to spread out. He will do this on his own initiative when he knows the conditions for subsidies and the special ecological relevance of bushes, weeds and rocks for faunal biodiversity. Here a lot of sensitizing activities has to accompany the implementation of decree.

Table 1. Some steps ameliorating the method considering the feed-backs of agricultural circles.

	Original version	Improved practicable version
Main criteria	Vegetation, fauna, few featured criteria	Vegetation, differentiated featured criteria
Key used to judge the ecological value of a pasture	Combining vegetation and features – combined subsidy	Separate evaluation of vegetation and structures – separate subsidies
Vegetation: indicative species	Original list with plants corresponding strongly with ecological quality	Adapted list by adding well known species and excluding species difficult to determine ( <i>Cirsium acaule</i> ), or prob-lematic in land use management ( <i>Cirsium</i> spp., <i>Cardus</i> spp.)
Problematic species like bracken and brambles	Brambles valued as appreciated thornbushes; low density of bracken accepted	Brambles not accepted as appreciated element. Definition of minimal surfaces. Bracken: pasture excluded when covering > 50%
Part of bushes in a pasture	Optimum: > 20%	Optimum: 10-25%

## Conclusions

In order to enhance biodiversity in agricultural areas by the responsibility of the farmers themselves, three presuppositions have to be satisfied:

- The scientific knowhow of the interconnections between vegetation, fauna, landscape features and further elements must be available and form the guideline in the promotional programme.
- The process how to develop and implement the promotional programme for practice must be worked out in a participatory way including stakeholders and experts. Agricultural extension is predestined to play the role of a facilitator in this process.
- The promotional programme must be broadly based in a political context and has to provide obvious economic incentives.

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# **An alternative paddock management system and its effect on grazing patterns and milk yield**

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## **Abstract**

Management methods have been shown to greatly synchronize grazing behaviour. Dairy cows can be expected to anticipate being moved to a fresh paddock after milking and synchronize their behaviour to this prediction. This results in reduction of grazing time and intake. This experiment was to test the effect of paddock management in reshaping behavioural patterns of cows to improve use of the pasture resource. Two control groups of cows were under typical half-day paddock management. Two treatment groups were moved to paddocks with distinct internal design. Half-day paddocks were fenced into two areas: main (85% of area) and remainder (15% of area). The main area was made available to cows when they arrived in a paddock. The remaining area of fresh forage was offered to them 3 hours before milking. Behavioural and production differences between treatment and control were tested using ANOVA. The magnitude of the difference was evaluated through Tukey-Kramer HSD. Treatment cows grazed 71 min d<sup>-1</sup> more, and had shorter rumination and leisure daily times ( $P < 0.01$ ) than controls. These behavioural adjustments happened only in afternoon and evening periods, when the fenced off pasture strip was released to the treatment group. Treatment cows produced 1.1 kg d<sup>-1</sup> more milk ( $P < 0.01$ ) than controls. A simple management practice such as this can greatly reshape cow grazing behaviour, and improve farm overall productivity and profitability.

Keywords: grazing management, grazing behaviour, pasture-based dairy, pasture management

## **Introduction**

Grazing is an example of a complex interrelationship among many biotic and abiotic ecosystem components. Management methods can greatly affect grazing behaviour adaptations (Taweel *et al.*, 2004). Dairy cows can be expected to anticipate being moved to a fresh paddock after milking, and synchronize their behaviour patterns to this prediction. This behaviour results in foraging reduction, especially around the reward time: 'Cows grazed very little towards the end of the period and waited for the next new area' (Orr *et al.*, 2001). This grazing delay was observed by many researchers (Holder, 1960; Balph and Balph, 1986; Olson and Richard, 1989; Barrett *et al.*, 2001), and was called 'waiting behaviour' (Schmitt, 1995). It was associated with low pasture intake (Arriaga-Jordan and Holmes, 1986; Orr *et al.*, 2001; Pulido and Leaver, 2003).

This study was designed to evaluate the effects of alternative paddock management as a tool to reshape dairy cow grazing habits to minimize waiting behaviour and increase use of pasture forage. Behavioural budgets and milk yield were used as parameters to test different paddock managements.

## **Materials and methods**

A herd of lactating Holstein cows was the subject of this trial at a commercial dairy farm in Hinesburg, Vermont, USA. The experimental herd grazed under Voisin management

intensive grazing (Murphy, 1998). Forty cows were paired and allocated to ten sets of four animals. Within each set, cows were randomly allocated to two treatments and two control groups, with ten animals in each group (Hernandez-Mendo and Leaver, 2006). Two control groups of cows were under typical half-day paddock management. Two treatment groups were moved to paddocks with distinct internal design. Half-day paddocks were fenced into two areas: main (85% of area) and remainder (15% of area). The main area was made available to cows when they arrived in a paddock. The remaining area of fresh forage was offered to them 3 hours before milking. Behavioural and production data were tested using ANOVA. The difference was evaluated through Tukey-Kramer HSD. Behavioural data from groups were used as unit of replication. Three days of behaviour measurements were used as means to avoid pseudo replication (Martin and Bateson, 1998).

## Results and discussion

Total grazing time during 24-hour periods was 71 min ( $P < 0.01$ ) longer for treatment cows than controls. Total grazing time was 498 min (SD = 47) for treatment, compared with 427 min (SD = 49,  $P < 0.01$ ) for control. These behavioural adjustments happened only in afternoon and evening periods, when the fenced-off pasture strip was released to the treatment group. Rumination and leisure time was affected negatively by alternative paddock management. Treatment paddock management had a positive effect on milk yield during the entire experiment. Treatment cows had an average daily milk yield of 1 kg higher than control cows. Treatment cows produced 20 kg cow<sup>-1</sup> (SD = 4), while control cows had a milk yield of 19 kg cow<sup>-1</sup> (SD = 4,  $P < 0.01$ ). This overall better performance of treatment cows resulted in greater milk yield in both June and August experimental trials. The June trial was characterized by an average daily milk yield of 20.4 kg cow<sup>-1</sup> (SD = 4.1), with a significant difference between control and treatment cows. Treatment cows produced 1.1 kg more than control cows during this trial ( $P < 0.01$ ). Treatment cows had an average daily yield of 20.9 kg cow<sup>-1</sup> (SD = 4.4), compared with 19.8 cow<sup>-1</sup> (SD = 3.8) for control cows. With an average daily milk yield of 18.5 kg cow<sup>-1</sup> (SD = 4.4), the August trial had a similar pattern. Treatment cows had an average daily milk yield 0.9 kg cow<sup>-1</sup> higher than control cows ( $P < 0.05$ ). Treatment cows produced 18.9 kg cow<sup>-1</sup> (SD = 4.4), while control cows produced 18.0 kg cow<sup>-1</sup> (SD = 4.4). The alternative paddock management discussed here was used to recondition feeding behaviour of cows by different moving schedules to distinct parts of paddocks. It motivated the herd to graze more, especially in the last hours of paddock occupation, thereby increasing total daily grazing, even under constant moving schedules. The higher performance of dairy cows under treatment management was a consequence of several behaviour adjustments to different environmental stimuli related to alternative paddock management. Treatment cows grazed longer and rumination and leisure times decreased, compared with the control cows. Milk yield of treatment cows increased, indicating that longer grazing time resulted in better utilization of the pasture resource, with less need for in-barn supplementation. Each cow under this alternative paddock management could produce a monthly increase in milk production of about 30 kg. At a current value to farmers of about \$0.33 per kg of milk, this would be worth \$10, which in most circumstances would justify the extra labour required.

## Conclusion

This study showed that simple farm management techniques can positively influence dairy cow performance and farm profitability, through environmental friendly use of pasture resources that otherwise would be wasted. Patient observation of livestock and plants is a

managerial skill that enables farmers to use management practices better integrated with the whole environment.

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# The seasonal biomass distribution of coprophagous beetles in fresh cow dung in a sub-mountain pasture

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## Abstract

The biomass of coprophagous beetles (*Scarabaeoidea*, *Hydrophilidae*) in fresh cow dung was monitored on a sub-mountain dairy farm using pitfall traps baited with 1.5 litre of dung. Major peaks of beetle activity were in the May, July and August / September sampling periods (mean dry weight biomass (DW) was 3.05 g, 3.52 g and 6.03 g trap<sup>-1</sup>, respectively), while in June and October beetle activity was lower (mean DW trap<sup>-1</sup>: 0.47 g, 0.51 g, respectively). In all samples the greatest biomass belonged to the species without burial breeding behaviour, whereas the tunnelling beetles (that lay eggs in dung buried in the soil) formed a minor part of sampled biomass (4–18% of mean DW trap<sup>-1</sup>). The highest impact of beetles on dung decomposition can be expected from August onwards due to larval feeding of the dung dweller *Aphodius rufipes*, contributing 80% of total DW biomass of the sample. The highest burial activity can be expected in May and in September due to breeding activity of *Ontophagus* spp. and *Geotrupes* spp., respectively.

Keywords: dung decomposition, coprophagous beetles, *Scarabaeoidea*, *Hydrophilidae*, cattle, pasture

## Introduction

The coprophagous beetles of *Scarabaeoidea* and *Hydrophilidae* form an important group of dung decomposers in pastures (Hanski and Cambefort, 1991). In the early phase of decomposition, beetles are the main agents of dung pat destruction. They increase the rate of dung disappearance and thereby improve the efficiency of pasture utilization (Holter, 1979; Gittings *et al.*, 1994). The species with burial-breeding activity (so-called tunnellers) contribute to dung pat disappearance by transporting pieces of dung into the soil in order to provide food source for their larvae. Nevertheless, all *Hydrophilidae* species and most *Scarabaeoidea* species in northern Europe do not express tunnelling behaviour (so-called dung dwellers). Dung pats are important for their adult feeding, mating and eventually breeding. The larvae contribute to dung disappearance more than adults in these species (Holter, 1979). The aim of this study was to survey the seasonal distribution of *Scarabaeoidea* and *Hydrophilidae* beetles on a sub-mountain cattle farm and to discuss their capacity for the dung decomposition.

## Materials and methods

The survey was carried out on a sub-mountain dairy farm (575 m a.s.l.) in southern Bohemia in the close vicinity of Natural Park of Novohradské Hory Mountains. Seasonal pasturage of approximately 100 cows of Czech Pied and Holstein breeds has been practised there for more than 30 years. The grazing generally starts in May and is abandoned in October. The area of pasture was 171 ha and it was covered by *Lolio-Cynosurenion* suballiance vegetation. The pasture area was surrounded by a landscape with forests, meadows, crop fields and small littorals. The beetles were caught in pitfall traps baited with 1.5 litre of fresh cow dung and

10% formaldehyde was used as a preserving fluid. A set of three traps was exposed in the pasture near the stables for 7 days in spring (Period 1: 19-26 May), in early summer (Period 2: 19-26 June), in mid- summer (Period 3: 24–31 July), in late summer (Period 4: 31 August-7 September) and in autumn (Period 5: 2–9 October). The collected beetles were identified to species (*Scarabaeoidea*) or to genera (*Hydrophilidae*) and divided into two groups according their breeding behaviour, the dung dwellers and the tunnellers. The dry body weight (DW) biomass of beetles was calculated using the mean weight of 11-36 specimens of particular species or genera (*Hydrophilidae*) dehydrated at 60°C for 72 hours.

## Results and discussion

The flight activity of beetles varied throughout the season (Table 1, Figure 1). The major peaks of beetle activity were in the May, July and August / September sampling periods (mean DW trap<sup>-1</sup>: 3.05 g, 3.52 g and 6.03 g, respectively), while in June and October periods beetle activity was lower (mean DW trap<sup>-1</sup>: 0.47 g, 0.51 g, respectively). The May peak was connected with flight activity of two small dung dwellers *Aphodius sphacelatus* and *Aphodius prodromus* (4-7 mm) contributing by 56% to a total DW biomass of the sample. While their larval development does not take place in dung (Gittings and Giller, 1997), their importance for dung pat decomposition is expected to be small. The July sample was formed from 72% of *Hydrophilidae*. Their larvae are carnivorous and do not contribute to dung decomposition (Hanski and Cambefort, 1991). The August / September peak was attributed to the occurrence of a large dung dweller *Aphodius rufipes* (11-13 mm) forming 80% of a total DW biomass of the sample. It occurred in traps from June, when adults of the new generation emerge (Gittings and Giller, 1997). Larvae feed on dung and their development to overwintering third instars can be accomplished in 6 weeks. This period of rapid growth is associated with massive food consumption, up to 300% of their dry body weight per day, and their feeding activity can be responsible for 14-20% of dung pat disappearance (Holter, 1979).

Table 1. The DW biomass of beetles collected in 5 sampling periods (the mean DW of three replicate traps). S: *Scarabaeoidea*; H: *Hydrophilidae*. deer and wild boar From a total of 19 species of *Scarabaeoidea* five species with tunnelling-nesting activity were identified in samples: *Onthophagus fracticornis*, *Onthophagus coenobita*, *Onthophagus joannae*, *Geotrupes spiniger* and *Geotrupes stercorarius*. Small tunnellers of *Onthophagus* spp. (most specimens appertained to *O. fracticornis*; 4.5-9 mm) create nests in spring up to 25 cm depth in soil. A higher impact on the destruction of dung pats can be expected by breeding activity of large *G. spiniger* and *G. stercorarius* (18-26 mm and 16-25 mm, respectively). They dig their nests up to 30 cm (*G. spiniger*) or 60 cm (*G. stercorarius*) beneath dung pat in September and in October (Balthasar, 1963).

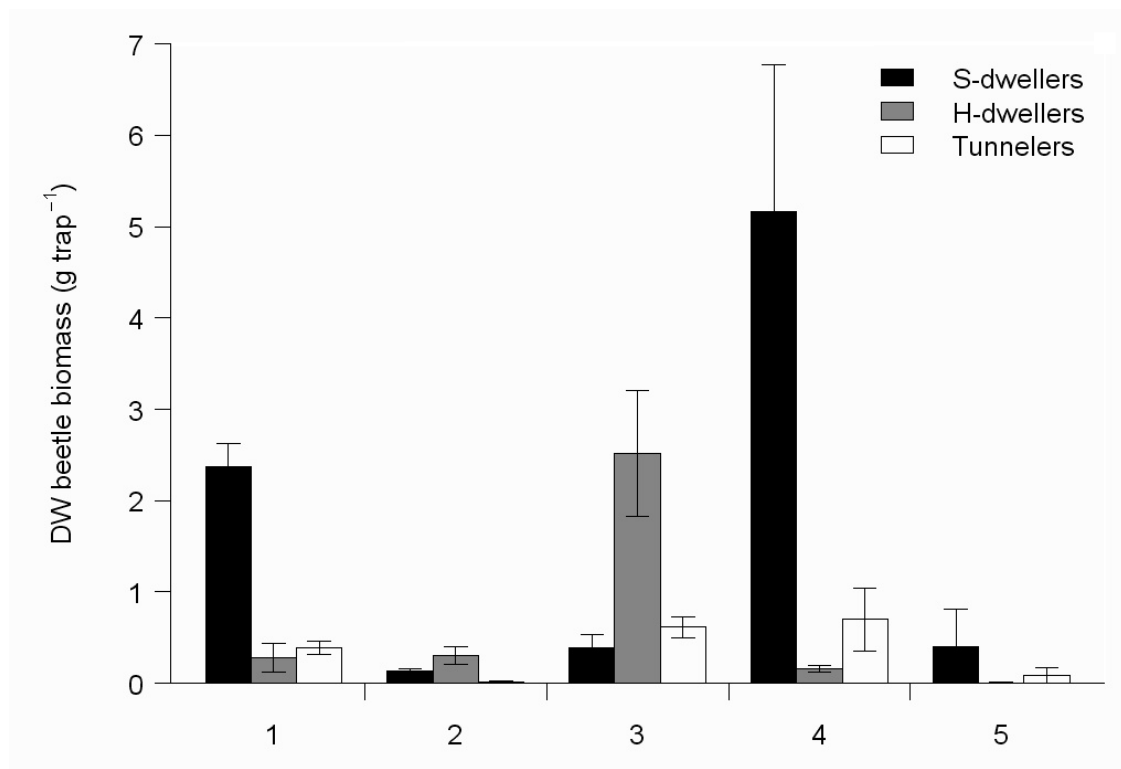


Figure 1. The DW biomass of beetles collected in five sampling periods (the means of three replicate traps and standard errors of the means are shown). S: *Scarabaeoidea*; H: *Hydrophilidae*.

## Conclusions

The greatest amount of caught beetle biomass belonged to the species without burial breeding behaviour, whereas the tunnelling beetles formed a minor part of sampled biomass (4-18% of mean DW trap<sup>-1</sup>). The highest impact of beetles on dung decomposition can be expected from August due to the larval feeding of a dung dweller *Aphodius rufipes*. The highest burial activity can be expected in May and in September due to the breeding activity of *Ontophagus* spp. and *Geotrupes* spp., respectively.

## Acknowledgement

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# Seed mixtures to establish species-rich meadows in the Swiss Central Plateau

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## Abstract

Since there is no appropriate seed bank in the soil, ecological compensation areas (ECAs) on permanent meadows seldom reach the desired species richness even though they are managed extensively (late cutting, no mineral fertilizer). This article summarizes experience gained in Switzerland of how this problem may be overcome by establishing new species-rich ECAs from seed mixtures. Four different species-rich mixtures were developed containing 32 to 47 species. According to the ecological sites where the mixtures are to be sown, their species compositions were adapted to the phytosociological associations *Arrhenatheretum elatioris* dry, *A. elatioris* moist, *Trisetetum flavescens* and *Mesobrometum erecti*. Several experiments and farm surveys have demonstrated that these mixtures are successful in generating species-rich ECAs. The species richness may even reach high ecological quality according to the 'Ecological Quality Ordinance', qualifying for additional payment. A highly specialized seed industry was built up to produce seed from local ecotypes. The number of species has multiplied and the area of seed production has increased greatly over the last 15 years. A key factor for this system's success was the transdisciplinary approach with close collaboration between research, the seed industry, farmers and nature conservation institutions.

Keywords: species-rich meadows, ecological compensation areas, wildflowers, seed industry, nature conservation

## Introduction

In the Swiss plateau species-rich meadows of the *Arrhenatheretum elatioris* association dominated the landscape until the middle of the 20<sup>th</sup> century. The increasing milk yield per cow and the availability of machinery and cheap fertilizer encouraged intensive grassland management. Consequently, species-rich meadows rapidly disappeared. In the new agricultural policy (Anonymous, 1992) Switzerland implemented ecological compensation areas (ECAs) as a means to increase biodiversity. The law requires that at least 7% of the agricultural utilization area of a farm has to be covered by ECAs, in order to receive direct payments. The vast majority of ECAs consist of extensively managed (late cutting, no mineral fertilizer) permanent meadows. However, most of these ECA meadows are of insufficient species richness (Hofer *et al.*, 2001) demonstrating that reduction of management intensity alone does not guarantee the transition into species-rich plant stands (Koch, 1996). The main reason is the lack of an appropriate seed bank in the soil following several decades of intensive management (Hald, 2004). Based on experience gained in Switzerland, this paper summarizes several aspects of overcoming this problem by seeding species-rich ECAs.

## Species composition of the seed mixtures

In order to keep the system transparent only four mixtures, each suitable for specific environmental conditions, were developed. The species composition of the mixtures (Table 1) was adapted to the phytosociological associations *Arrhenatheretum elatioris* dry, *A. elatioris* moist, *Trisetetum flavescens* and *Mesobrometum erecti*. The mixtures consist of 9-10 grass species, 5-9 legumes and 13-29 forbs (wildflowers). Total species number is between 32

Table 1. Mixtures for ECA meadows: overview of their composition.

Type of mixture	No. of grasses	Differential grass species	No. of dicots	Example of typical dicot species	No. of species for ecol. quality
<i>Arrhenatheretum</i> (dry)	10	<i>Arrhenatherum elatius</i>	28	<i>Centaurea scabiosa</i> , <i>Salvia pratensis</i> , <i>Knautia arvensis</i>	24
<i>Arrhenatheretum</i> (moist)	10	<i>Arrhenatherum elatius</i> , <i>Agrostis gigantea</i> , <i>Alopecurus pratensis</i>	22	<i>Sanguisorba officinalis</i> , <i>Lychnis flos-cuculi</i> , <i>Cirsium oleraceum</i>	14
<i>Trisetetum</i>	10	<i>Agrostis capillaris</i> , <i>Cynosurus cristatus</i>	23	<i>Silene vulgaris</i> , <i>S. dioica</i> , <i>Prunella vulgaris</i>	15
<i>Mesobrometum</i>	9	<i>Bromus erectus</i> , <i>Koeleria pyramidata</i>	38	<i>Hieracium pilosella</i> , <i>Stachys officinalis</i> , <i>Hippocrepis comosa</i>	33

Common species: *Trisetum flavescens*, *Festuca rubra*, *F. pratensis*, *Poa pratensis*, *Helictotrichon pubescens*, *Anthoxanthum odoratum*, *Lotus corniculatus*, *Medicago lupulina*, *Trifolium pratense*, *Lathyrus pratensis*, *Centaurea jacea*, *Ajuga reptans*, *Leontodon hispidus*, *Leucanthemum vulgare*, *Plantago lanceolata*, *Tragopogon orientalis*

and 47. Besides a set of species that is common to all four mixtures there are differentiating species typical for the respective phytosociological association (Table 1; Suter *et al.*, 2004). The mixtures were developed in close cooperation with the Swiss Commission for Wild Plant Conservation (CPS/SKEW) to help prevent distortion of the typical local flora. Thus, only species that are found over the whole biogeographical region where the sowing is to take place (Swiss Central Plateau including the Jura-Mountains and northern slopes of the Alps) were introduced into the seed mixtures. Species that occur only in sub-regions were excluded.

### Successful establishment of new species-rich meadows

The establishment of new species-rich meadows was tested with different variations of species-rich seed mixtures in over 20 medium- (3 years) to long-term (17 years) experiments at more than 40 sites. While success was poor at the beginning, the adaptation of the seed mixture composition increased the success rate considerably (Jacot and Lehmann, 2001). An assessment of more than 40 sown ECA meadows from plot experiments revealed that in approximately 50% , more than two-thirds of the sown species established, and in about 95% it was more than half (data not shown). These outstanding results were confirmed by an experimental series conducted on-farm at 380 sites where different mixtures and sowing methods were tested in practice (Steinegger and Koch, 1997).

The Ecological Quality Ordinance (Anonymous, 2001) introduced additional payment for ECAs with high richness of valuable species in Switzerland. To meet this criterion an ECA must have at least 6 indicator species for ecological quality per 25 m<sup>2</sup>. Since the species-rich seed mixtures contain between 14 and 24 of the indicator species listed in the ordinance (Table 1) their potential to generate ECAs with high ecological quality is considerable.

### Production and sales of high quality seed

The area of wildflower seed production increased greatly between 1991 and 2006 (Figure 1) and the number of species with commercial seed available reached 50 in 2001 (data not shown). The labour-intensive production of wildflower seed includes the collection of various ecotypes, sowing in pots and growing the seedlings in a greenhouse before planting them in the field, harvesting seed several times due to uneven maturation phases of many plant species and then gently drying the harvested seeds. Collecting ecotypes has to be conducted according to the guidelines of the Swiss Commission for Wild Plant Conservation (CPS/SKEW) and is supervised by specialists. To preserve genetic variability, the plants used for seed production



**Seed production of wildflowers**

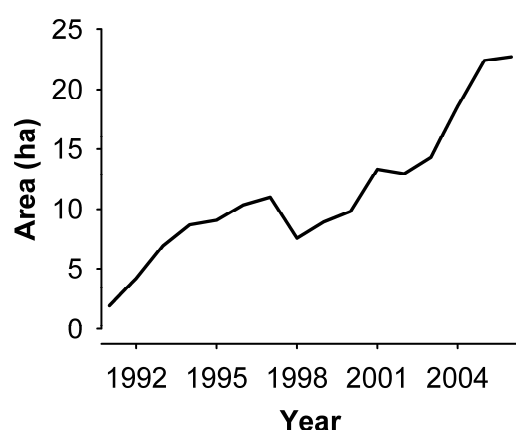


Figure 1. Production of wildflower seed.

**Seed sales in area equivalents**

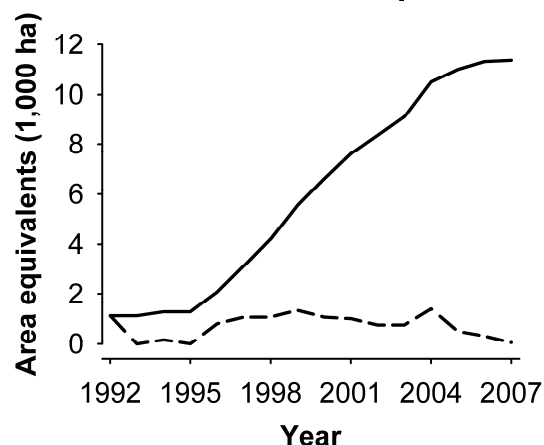


Figure 2. Species-rich meadows: annual (dashed) and cumulative (solid) sales.

have to be replaced regularly by newly collected material. To prevent hybridization, outbreeding species have to be spatially separated. The species purity and weed percentage of all the production fields is inspected by officials of the Swiss Seed Quality Laboratory. In addition, seed samples from at least 25% of the production fields are tested for purity and germination ability by the Swiss Seed Quality Laboratory.

To encourage the acceptance of species-rich seed mixtures with wildflower species, the composition was adapted to keep the price at a reasonable level. The amount of seed from very expensive species was reduced, to keep the price of the seed mixtures below CHF 1,300 per hectare. The moderate prices and the introduction of an extra payment (CHF 500 per ha per year) for ECAs with a high richness in valuable species, resulted in seed being sold that corresponded to 11,000 hectares (Figure 2) from 1992 to 2007. This area represents more than 10% of the ECA meadow area.

## Conclusion

A key factor for the system's success was the transdisciplinary approach with a close collaboration between research, seed industry, farmers and nature conservation institutions.

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# Does the effect of selection history influence vegetation and plant preferences by cattle?

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## Abstract

In this project plant and vegetation preferences of two Norwegian dairy cattle breeds with different selection history were studied. The Norwegian dairy breed Black-sided Trønder and Nordland Cattle (STN) has never been selected efficiently for higher milk production. The other breed, however, the Norwegian Red (NR), has mainly been selected for this. Two herds, both consisting of STN and NR cows, were studied. To examine the animals' plant preferences, faeces samples were collected and analysed for plant fragments. Vegetation maps were also used to find possible differences in grazing preferences. Breed differences, with regard to recorded plant fragments in the faeces samples, were significant for *Nardus stricta*, a species characteristic of nutrient-poor but mostly species-rich vegetation types in the studied areas; these vegetation types of high importance for biodiversity especially in one of the areas. STN had the highest share of *Nardus stricta*. The results of the study indicate that a higher producing cattle breed might prefer to graze more nutrient-rich vegetation areas compared to a lower yielding cattle breed, when grazing less-nutrient and base-rich, but species-rich, grasslands.

Keywords: semi-natural grasslands, grazing, cattle, breed differences, vegetation preferences

## Introduction

The areas of species-rich semi-natural grasslands have been considerably reduced during the last 50 years in Norway. Maintenance of these valuable habitats depends on traditional use or special management to prevent regrowth succession and loss of species (Norderhaug *et al.*, 1999; Sickel *et al.*, 2004). A survey conducted by Sæther and Vangen (2001) revealed that farmers utilizing the low yielding Norwegian dairy breed Black-sided Trønder and Nordland Cattle (STN) expected this breed to utilize extensive grasslands better than cows of the high yielding Norwegian Red (NR). Approximately 97% of Norwegian dairy cattle are NR, while 0.1% belongs to the STN breed. Average yearly milk yield for the two breeds are 6,200 kg and 4,000 kg, respectively. Several studies have shown that breeds and/or lines selected for high production intensity generally show lower levels of activity (references in Sæther *et al.*, 2006). Furthermore, when given the choice between hardly accessible and easily accessible fodder, they tend to choose easily accessible fodder at a higher rate than breeds and lines not selected for the same high production intensity. This study therefore addresses the following question: Will genetically high yielding cows focus more on nutrient-rich plant species and vegetation types than genetically low yielding cows when grazing extensive, species-rich semi-natural grasslands?

## Materials and methods

Two dairy cattle herds, one farm in Skåbu and one farm in Valdres, were studied. Both farms have long, continuous summer farming traditions based on semi-natural grasslands in the mountains. The herds had nine pure-bred cows each, five and six STN cows, and four and three NR cows, respectively. In Skåbu the dominating bedrocks are base-rich with a high weathering capacity, giving rise to nutrient- and base-rich soils. In Valdres the bedrock consists entirely of a less basic phyllite type, and the soils are poorer than in Skåbu. Today the studied areas have a low grazing pressure. On average the herds on each farm were observed 9 hours daily, with a span of 7-13 hours per day, during one week in July and one in August. In total, 49 faeces samples were randomly collected across weeks, study sites and animals and prepared for microhistological analyses, following the procedures of Garcia-Gonzalez (1986). Fragments from 28 plant-species, -genera and -groups, were found in the faeces samples; however, only the 14 with mean values > 2% of total recorded fragments are included in the statistical analyses. In the statistical analysis the possible effects of breed (STN and NR), farm (Skåbu and Valdres) and month (July and August) are included as fixed effects, together with the interaction breed x farm. The effect of cow within breed is included as a random effect to adjust for variation between individuals within breed. The nomenclature for plants follows Lid and Lid (2005).

## Results and discussion

Both study sites had species-rich grasslands. In total, 123 species were found. Of these, 51 species were only found in Skåbu and 20 species only in Valdres. The most common grassland vegetation type in Valdres was characterized by many common and small-sized grasses and herbs with low to medium demands of base-rich soil. *Nardus stricta* and other drought-resistant species often grew on the top of low elevations, while species indicating better moisture conditions, e.g. certain *Carex* species, dominated on lower and more humid parts of the grasslands. *Vaccinium myrtillus* was also often found here. Another grassland type found in the Valdres area is dominated by *Nardus stricta* as a result of too heavy grazing for many years. The vegetation in Skåbu was characterized by many base-demanding species. Forests, protecting small elevations and knolls against wind and drought, surrounded the grasslands. Thus, moisture conditions and the distribution of plant species in the grasslands were more uniform in these situations.

Table 1. Results from the statistical analyses on the plant fragment residues observed in the faeces samples. The figures are given as percentage of total fragments.

Plant species, genera or group	Farm	Breed	Month	Interaction Farm/breed	Average value	LS-means STN	LS-means NR	LS-means Skåbu	LS-means Valdres
<i>Vaccinium myrtillus</i>	**	-	-	*	2.0	1.8	2.1	2.8	1.3
<i>Nardus stricta</i>	**	*	-	-	3.8	5.1	3.8	1.7	7.2
<i>Carex</i> spp	**	-	-	-	8.4	8.4	9.5	4.4	13.5
Total grass	**	-	-	**	76.1	77.7	75.8	74.0	79.4
<i>Deschampsia cespitosa</i>	**	-	-	-	25.5	27.0	25.1	19.3	32.9
<i>Avenella flexuosa</i>	**	-	**	-	15.1	15.0	14.9	19.8	10.2

The results from the microhistological analyses showed that the faeces of cows contained mostly grass fragments (76%). In addition to a group of unidentified grass species, 13 grass species and genera were recognized. *Avenella flexuosa* (25%) and *Deschampsia cespitosa* (15%) dominated. Also, several herbs and *Carex* spp. were valuable grazing plants. Together herbs and *Carex* spp. constituted almost 16% of the observed fragments. Only the dominating

grazed grass species, and the species or plant groups contributing significantly to the interpretation of the results, are included in the statistical analysis presented below.

Table 1 shows that the effect of breed was only significant for *Nardus stricta*, a species characteristic of nutrient-poor but mostly species-rich vegetation types in the studied areas. The STN breed had a higher share of *Nardus stricta* fragments in the faeces samples than the NR breed. The interaction between farm and breed was significant for *Vaccinium myrtillus* and the plant group “total grass”. The effect of farm was significant for all tested species and plant groups; however, within farm the results showed that when grazing in areas with nutrient- and base-rich soil and species-rich vegetation types, as in Skåbu, the two breeds mostly grazed the same vegetation and plant species. In Valdres, however, where the soil is less fertile, and the plant species distribution less uniform than in Skåbu, the results indicate that the NR cows grazed more in patches where *Carex* spp. grow than did the STN cows. STN also grazed more *Carex* spp. in Valdres than in Skåbu, but the increase was less than for NR. Both breeds grazed less *Vaccinium myrtillus* in Valdres, but NR decreased less than STN. In Valdres *Vaccinium myrtillus* occurs in the same vegetation type as some *Carex* spp. that are defined as valuable grazing plants (Rekdal, 2001). Studies by Garmo (1986) state that *Carex* spp. have a higher content of crude protein and less content of crude fibre than grass species during the entire grazing season, indicating that *Carex* spp. in general have a higher nutrient value than grass. This may explain why NR cows seem to graze more in patches where *Carex* spp. grow and *Vaccinium myrtillus* occur. Both breeds grazed more *Nardus stricta* in Valdres than in Skåbu, probably because this species is more common in Valdres. This grass species has low digestibility and stiff, narrow leaves. It is therefore not seen as a valuable or preferred grazing species. It may, however, be grazed to a certain extent when it occurs in vegetation types preferred by the grazing cattle. STN grazing resulted in a greater increase in *Nardus stricta* than NR, indicating that the STN cows graze more in nutrient-poor but species-rich areas where this species is common in Valdres.

## Conclusions

Results from this study indicate that a cattle breed selected for high milk yield might prefer to graze more nutrient-rich vegetation areas, compared with a lower yielding cattle breed, when grazing less nutrient- and base-rich but species-rich grasslands. When managing semi-natural grasslands, the effect of grazing, therefore, might differ between low- and high-yielding dairy cattle breeds. This possible difference ought to be taken into consideration when managing extensive grasslands, as loss of biodiversity by regrowth of less nutrient-rich but species-rich vegetation types seems to be smaller when using a lower yielding breed.

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# Long-term effects of mulching on botanical composition, yield and nutrient budget of permanent grassland

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## Abstract

Mulching is an alternative management for permanent grassland currently not in agricultural production. To test the hypothesis that mulching leads to nutrient enrichment, loss of biodiversity and dominance of grasses, a two-factorial experiment was run from 1987 to 2004. Mulching (M) was compared with cutting and removal of the biomass, both with (CF) and without fertilization (C) at five defoliation frequencies on an improved meadow in southwest Germany. Botanical composition, biomass yield, nutrient removal and soil nutrient content were recorded. Except at the highest defoliation frequency, biomass yields of the M treatment were as high as those of the CF treatment. Under both treatments, yields increased during the experimental period, indicating that nutrient enrichment took place. Species number at the end of the experiment was not different between the M and C treatments, but was lower under the CF treatment. Yield percentage of grasses was not increased by mulching in comparison with cutting and removing of biomass. Defoliation frequency was the major factor influencing botanical composition. At only one or two defoliations per year, all three treatments resulted in species-poor, grass-rich swards.

Keywords: mulching, defoliation frequency, biodiversity, nutrient budget, landscape preservation

## Introduction

Mulching, as an alternative management for areas of permanent grassland not presently needed to provide forage, has been discussed controversially (Spatz, 1994; Laser 2002). As the minimum management of permanent grassland under Cross Compliance in Germany is met by mulching once a year, a widespread adoption of this practice may occur. Since biomass is not removed, mulching may lead to nutrient accumulation. Moreover, it may favour grasses at the expense of forbs, which are less capable of penetrating a thick layer of mulch, thus leading to a loss of biodiversity. The aim of this study was to test these hypotheses at different defoliation frequencies in a long-term study.

## Materials and methods

A two-factorial experiment, laid out as a split-plot design with two replications, was run from 1987 to 2004 on a stagnic luvisol at the experimental station Aulendorf. This site is situated in the alpine foothills of southwest Germany at 590 m asl; average yearly precipitation is 910 mm and average yearly temperature is 7.9°C. Before the start of the experiment, the grassland, an improved meadow, had been cut four times a year and fertilized with liquid manure. As a first experimental factor, mulching (M) was compared with cutting and removal of biomass without fertilizer application (C) and with cutting and removal with fertilizer application (CF). For the CF treatment, fertilizer was applied in the form of calcium ammonium nitrate, rock phosphate, muriate of potash and kieserite. Fertilizer levels were chosen to balance the expected nutrient removal in the harvested biomass. As a second factor, the defoliation frequency was varied from one to five defoliations per year. Defoliation dates

and fertilizer levels in the CF treatment for each defoliation frequency are listed in Table 1. Botanical composition was recorded every one to two years by estimating the proportion of total dry matter yield for all vascular plant species on each 5.6 x 2.7-m plot. Biomass samples were taken at each defoliation and analysed for N, P, and K content. Laboratory analyses followed the methods of the VDLUFA (2002). ANOVA was performed on biomass yield, species number and yield percentage of grasses using the procedure MIXED of SAS System 9.1. To assess development of biomass yields over time, differences were calculated between each observed yield data point and the average yield of the respective experimental plot over the whole period of the experiment. This period was then divided into three sub-periods of 6 years each and subjected to ANOVA. In each case, means were compared using HSD.

Table 1. Defoliation dates and fertilizer levels in the experiment.

Defoliations [ $y^{-1}$ ]	Defoliation dates (M, C, CF)	Fertilizer input [ $kg\ ha^{-1}\ y^{-1}$ ] (CF only)
1	01.09.	N: 100 / P: 17 / K: 100 / Mg: 13
2	20.07. / 20.10.	N: 150 / P: 26 / K: 149 / Mg: 18
3	01.06. / 20.07. / 20.10.	N: 200 / P: 35 / K: 198 / Mg: 24
4	15.05. / 15.06. / 20.07. / 20.10.	N: 250 / P: 44 / K: 248 / Mg: 30
5	15.05. / 15.06. / 20.07. / 01.09. / 20.10.	N: 320 / P: 57 / K: 314 / Mg: 37

## Results and discussion

Biomass yields of the mulching treatment were significantly lower than those of the fertilized cutting treatment only at the highest defoliation frequency. In contrast, they were significantly higher than those of the unfertilized cutting treatment except the lowest defoliation frequency (Table 1). At all defoliation frequencies, biomass yields during the last six years of the experiment were significantly higher than the average yield during the whole period for the CF and M treatment, while they were significantly lower than average in the C treatment. (Figure1). As biomass yields are an indicator of site fertility, this indicates that in the unfertilized cutting treatment nutrient impoverishment took place, while in the mulching treatment and, to a greater extent, in the fertilized cut treatment, nutrient enrichment occurred.

Table 2. Biomass yields [ $Mg\ DM\ ha^{-1}\ y^{-1}$ ] ( $\pm$  SE) in the three treatments at five defoliation frequencies. Different lower case letters denote statistically different means within a column, different capital letters statistically different means within a row ( $P = 0.05$ ).

Defoliations [ $y^{-1}$ ]	Cutting without fertilizer (C)	Mulching (M)	Cutting with fertilizer (CF)
1	5.04 $\pm$ 0.16 b A	6.27 $\pm$ 0.27 c A	6.34 $\pm$ 0.22 c A
2	6.38 $\pm$ 0.18 a B	8.46 $\pm$ 0.24 ab A	9.83 $\pm$ 0.29 b A
3	5.99 $\pm$ 0.26 ab B	10.56 $\pm$ 0.30a A	12.29 $\pm$ 0.41 a A
4	5.15 $\pm$ 0.25 b B	9.06 $\pm$ 0.28 ab A	11.06 $\pm$ 0.31 ab A
5	5.79 $\pm$ 0.30 ab C	8.10 $\pm$ 0.30 b B	12.58 $\pm$ 0.36 a A

At one defoliation per year, N, P and K removal in the M treatment was 89, 98 and 83%, respectively, of the nutrient removal in the CF treatment, indicating that especially for N and K higher losses occurred when nutrients were returned in the form of mulched biomass instead of mineral fertilizer. The losses increased with increasing defoliation frequency, as at five defoliations per year, N, P and K removal in the M treatment was only 63, 68 and 45%, respectively, of the nutrient removal in the CF treatment. This may be attributed to the faster degradation of less mature mulched biomass at high defoliation frequencies. At the end of the experiment yield percentage of grasses was high at all defoliation levels in the CF treatment, while in the M treatment, and even more so in the C treatment, the yield percentage of forbs and legumes increased with increasing defoliation frequency (Table 3). Mean species number per plot was not different between M and C treatment (19.6 and 20.8 species, respectively),

but was significantly lower in the CF treatment (15.4 species). In all treatments, it increased significantly from one or two defoliations (13.0 and 14.7 species) to three to five defoliations (21.9, 21.0, and 22.4 species). This unusual response can be attributed to the fact that the site had been managed at a high intensity before the experiment was started, which is still the case with the grassland in the vicinity of the experimental site. As a result, even after a period of seventeen years hardly any species adapted to low utilization frequency had established at the experimental site, and grasses like *Arrhenatherum elatius* and *Holcus lanatus* dominated on the plots with low defoliation frequency.

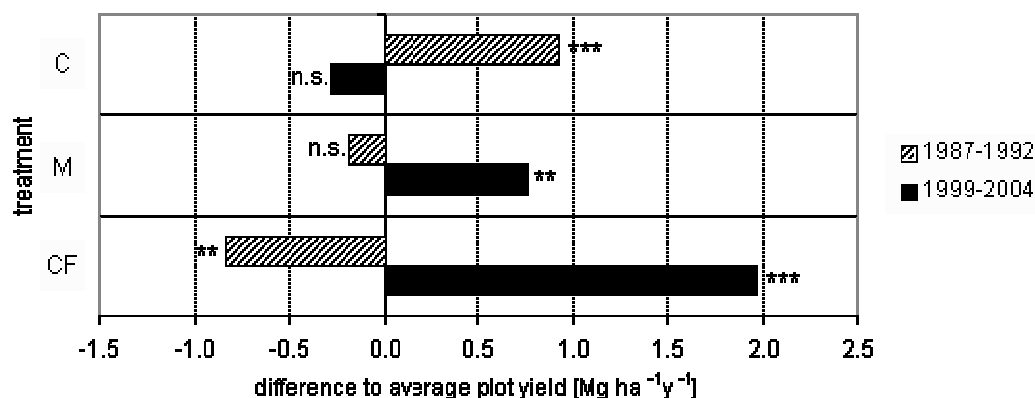


Figure 1. Yield difference of the first (1987-1992) and the last (1999-2004) sub-period to the average yield over the whole duration of the experiment (1987-2004). Symbols indicate if the values differ significantly from zero.

Table 3. Yield percentage of grasses ( $\pm$  SE) in the three treatments at five defoliation frequencies (1998-2004). Different lower case letters denote significant differences within a column, different capital letters significant differences within a row ( $P = 0.05$ ).

Defoliations [ $y^{-1}$ ]	Cutting without fertilizer (C)	Mulching (M)	Cutting with fertilizer (CF)
1	83.7 $\pm$ 1.2 a A	95.5 $\pm$ 0.6 a A	97.8 $\pm$ 0.1 a A
2	71.7 $\pm$ 1.3 ab B	92.2 $\pm$ 1.9 ab AB	95.0 $\pm$ 0.6 a A
3	50.0 $\pm$ 2.1 b B	71.7 $\pm$ 2.8 bc AB	90.0 $\pm$ 0.5 a A
4	38.3 $\pm$ 2.5 c B	58.3 $\pm$ 6.1 c B	93.8 $\pm$ 0.5 a A
5	45.0 $\pm$ 4.2 bc B	54.2 $\pm$ 3.0 c B	87.8 $\pm$ 1.5 a A

## Conclusions

Observed yields and calculated nutrient efficiencies indicate that a large proportion of the nutrients returned to the soil by mulched plant material is available for plant growth, and that in the course of seventeen years nutrient enrichment occurred on the mulched plots of the experimental site. On this originally species-poor grassland mulching had no adverse effect on the plant composition. Particularly at higher mulching frequencies species number and percentage of forbs and legumes approached those observed at the unfertilized cutting treatment. However, at only one defoliation per year, which corresponds to the minimum management under Cross Compliance, species number decreased and yield percentage of grasses increased both under mulching and cutting with removal.

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# The influence of grassland management on biodiversity in the mountainous region of NE Romania

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## Abstract

The permanent grasslands of north-eastern Romania, situated on moderate natural fertility soils, are generally of low productivity and have a botanical composition that produces fodder of low quality. This paper presents results of a study of the influence of grassland management in mountainous north-eastern Romania on phytocoenotic biodiversity and rate of species change. We studied the effect on botanical composition of fertilization using 100-200 kg N ha<sup>-1</sup> + 100-200 kg P ha<sup>-1</sup> on a *Nardus stricta* grassland at Cosna-Suceava, and of 10-30 Mg ha<sup>-1</sup> manure fertilization on a background of 30-50 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>, on an *Agrostis capillaris* + *Festuca rubra* grassland at Dorna Arini-Suceava (both sites from the intra-mountainous Depression of Vatra Dornei). The high mineral fertilizer rates resulted in a change of the dominant species in *N. stricta* grassland, increasing the percentage of *F. rubra* and *A. capillaris* from 3-5% to 20-55%, while the organic and mineral fertilization on *A. capillaris* + *F. rubra* grassland resulted in increasing the percentage of *Cynosurus cristatus* from 7% to 15-36%.

Keywords: biodiversity, fertilization, mountainous grasslands

## Introduction

The problem of biodiversity is one of the main international concerns because human activities generally, and intensive agriculture in particular, endanger the integrity of the spontaneous gene pool. The goal of intensive agricultural methods is to get higher yields, rather than obtain long-term effects on quality and diversity of natural vegetation. The experience of developed countries indicates that making decisions on biodiversity must be done only after carrying out in-depth studies, based on scientific results, which allow a sustainable management of natural resources, among which permanent grasslands have an important place (Hopkins *et al.*, 1999; Mendarte *et al.*, 2005; Poetsch *et al.*, 2005; Vintu *et al.*, 2007).

In Romania, about half of the > 4.9 million ha of permanent grasslands are found in the mountainous region. As they have great floristic diversity, studies are required which involve identifying the means of controlling the pasture area without risking the phytocoenotic biodiversity, but efficient enough to satisfy the needs of animal breeders.

## Materials and methods

In order to determine the vegetation from the area under study, we used the geobotanical method, biodiversity being studied by means of floristic sampling at two sites in the intra-mountainous Depression of Vatra Dornei, Cosna and Dorna Arini (820-840 m height asl, and 672-756 mm annual rainfall) in the period 2006-2007. For identifying changes in the grassland vegetation, two trials were set up in the period 2006-2007, one with mineral fertilization and another with mineral and organic fertilization. The vegetation was studied by the geobotanical method by means of a 25 m<sup>2</sup> framework for vegetation, which requires



species recording from canopies and their specific covering. On the *Nardus stricta* grassland at Cosna-Suceava, six variants of mineral fertilization were used: V<sub>1</sub> – control (unfertilized), V<sub>2</sub> – 100 kg N ha<sup>-1</sup> + 100 kg P ha<sup>-1</sup>, V<sub>3</sub> – 140 kg N ha<sup>-1</sup> + 140 kg P ha<sup>-1</sup>, V<sub>4</sub> – 200 kg N ha<sup>-1</sup> + 200 kg P ha<sup>-1</sup>, V<sub>5</sub> – 100 kg N ha<sup>-1</sup> + 100 kg P ha<sup>-1</sup> and 40 kg N ha<sup>-1</sup> + 40 kg N ha<sup>-1</sup> (after the first cycle), V<sub>6</sub> – 100 kg N ha<sup>-1</sup> + 100 kg P ha<sup>-1</sup> and 100 kg N ha<sup>-1</sup> + 100 kg P ha<sup>-1</sup> (after the first cycle), V<sub>7</sub> – 80 kg N ha<sup>-1</sup> + 80 kg P ha<sup>-1</sup> and 60 kg N ha<sup>-1</sup> + 60 kg P ha<sup>-1</sup> (after the first cycle). On the *Agrostis capillaris* + *Festuca rubra* grassland at Dorna Arini-Suceava, six variants of organic and mineral fertilization were used: V<sub>1</sub> – control (unfertilized), V<sub>2</sub> – 10 Mg ha<sup>-1</sup> cattle manure, applied each year + 30 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>, V<sub>3</sub> – 10 Mg ha<sup>-1</sup> cattle manure, applied each year and 30 + 20 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>, V<sub>4</sub> – 20 Mg ha<sup>-1</sup> cattle manure, applied every two years and 30 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>, V<sub>5</sub> – 20 Mg ha<sup>-1</sup> cattle manure, applied every two years and 30 + 20 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>, V<sub>6</sub> – 30 Mg ha<sup>-1</sup> cattle manure, applied every three years and 30 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>, V<sub>7</sub> – 30 Mg ha<sup>-1</sup> cattle manure, every three years and 30 + 20 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>. Harvesting was done as a hayfield crop, at the ear formation of dominant grasses, and determinations were carried out at the first vegetation cycle.

## Results and discussion

On *Agrostis capillaris* and *Festurubra* grassland, 38 species were found, of which 13 were grasses, 3 legumes and 22 species from other botanical families; the dominant species were *Agrostis capillaris* (35%) and *Festuca rubra* (18%), followed by *Nardus stricta*, *Cynosurus cristatus* and *Alchemilla vulgaris* (Table 1).

Table 1. Influence of fertilization on biodiversity and percentage of species in the canopy (%).

Species	<i>Agrostis capillaris</i> and <i>Festuca rubra</i>							<i>Nardus stricta</i>						
	V <sub>1</sub>	V <sub>2</sub>	V <sub>3</sub>	V <sub>4</sub>	V <sub>5</sub>	V <sub>6</sub>	V <sub>7</sub>	V <sub>1</sub>	V <sub>2</sub>	V <sub>3</sub>	V <sub>4</sub>	V <sub>5</sub>	V <sub>6</sub>	V <sub>7</sub>
<i>Agrostis capillaris</i>	35.0	49.0	51.0	21.0	30.0	24.0	22.0	3	25	35	45	21	55	47
<i>Anthoxanthum odoratum</i>	3.0	1.0	2.0	3.0	4.0	4.0	4.0	3	2	2	5	4	5	3
<i>Briza media</i>	+	+	+	+	1.0	1.0	4.0	2	4	2	7	2	2	2
<i>Cynosurus cristatus</i>	7.0	15.0	17.0	19.0	24.0	29.0	36.0	1	2	+	2	+	+	1
<i>Dactylis glomerata</i>	+	-	-	-	-	-	-	-	-	-	+	-	-	-
<i>Deschampsia caespitosa</i>	-	+	-	-	2.0	2.0	1.0	-	-	-	-	-	-	-
<i>Festuca pratensis</i>	+	+	-	2.0	1.0	-	2.0	-	-	+	-	+	-	+
<i>Festuca rubra</i>	18.0	5.0	4.0	8.0	6.0	5.0	4.0	5	16	25	25	40	20	20
<i>Holcus lanatus</i>	3.0	3.0	2.0	3.0	3.0	2.0	6.0	-	-	-	+	-	-	-
<i>Nardus stricta</i>	13.0	10.0	7.0	5.0	8.0	9.0	7.0	76	32	25	9	24	11	9
<i>Phleum pratense</i>	-	+	-	-	+	+	+	-	-	-	-	-	-	-
<i>Poa trivialis</i>	-	1.0	1.0	4.0	+	+	+	-	-	-	-	-	-	-
<i>Trisetum flavescens</i>	1.0	7.0	2.0	1.0	1.0	+	+	-	-	-	-	-	-	-
<b>Grasses</b>	<b>80.0</b>	<b>91.0</b>	<b>86.0</b>	<b>66.0</b>	<b>79.0</b>	<b>76.0</b>	<b>86.0</b>	<b>81</b>	<b>89</b>	<b>93</b>	<b>91</b>	<b>93</b>	<b>82</b>	
<i>Trifolium pratense</i>	3.0	1.0	1.0	1.0	2.0	4.0	2.0	1	2	2	1	1	1	4
<i>Trifolium repens</i>	2.0	1.0	1.0	3.0	6.0	7.0	4.0	1	8	-	-	1	1	2
<i>Lotus corniculatus</i>	+	-	-	-	-	-	-	-	-	3	1	-	-	2
<b>Legumes</b>	<b>5.0</b>	<b>2.0</b>	<b>2.0</b>	<b>4.0</b>	<b>8.0</b>	<b>11.0</b>	<b>6.0</b>	<b>10</b>	<b>5</b>	<b>2</b>	<b>2</b>	<b>2</b>	<b>8</b>	
<i>Achillea millefolium</i>	2.0	+	0.5	0.5	+	+	+	+	+	+	1	1	+	5
<i>Alchemilla vulgaris</i>	8.0	5.0	5.0	8.0	7.0	9.0	6.0	1	+	+	-	+	1	-
<i>Chrysanthemum leucanthemum</i>	1.0	1.0	+	+	+	0.5	+	+	+	-	-	-	+	+
<i>Cruciatia glabra</i>	0.5	+	+	+	-	+	-	+	+	-	+	+	+	-
<i>Plantago lanceolata</i>	0.5	0.5	1.0	4.0	1.0	+	1.0	+	+	+	+	+	+	+
<i>Potentilla erecta</i>	2.0	+	+	+	0.5	+	+	1	1	2	1	1	+	+
<i>Prunella vulgaris</i>	+	0.5	0.5	+	+	+	+	1	+	+	+	+	-	+
<i>Ranunculus acer</i>	+	+	+	1.0	1.5	3.0	1.0	+	+	+	+	+	+	+
<i>Taraxacum officinalis</i>	+	+	-	1.0	1.0	+	+	+	+	+	+	+	+	+
<i>Thymus pulegioides</i>	-	-	-	-	-	-	-	3	3	1	1	+	+	2
<i>Veronica arvensis</i>	+	+	+	+	-	-	-	-	-	-	-	-	-	-
<i>Other species</i>	+	+	+	+	+	+	+	+	+	+	+	+	+	+
<b>Forbs</b>	<b>15.0</b>	<b>7.0</b>	<b>7.0</b>	<b>15.0</b>	<b>13.0</b>	<b>13.0</b>	<b>8.0</b>	<b>8</b>	<b>7</b>	<b>6</b>	<b>5</b>	<b>7</b>	<b>5</b>	<b>10</b>
<b>Covering %</b>	<b>100</b>	<b>100</b>	<b>95</b>	<b>85</b>	<b>100</b>	<b>100</b>	<b>100</b>	<b>100</b>	<b>98</b>	<b>100</b>	<b>100</b>	<b>100</b>	<b>100</b>	<b>100</b>
<b>Blanks %</b>	<b>0.0</b>	<b>0.0</b>	<b>5.0</b>	<b>15.0</b>	<b>0.0</b>	<b>0.0</b>	<b>0.0</b>	<b>0</b>	<b>2</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>	<b>0</b>
<b>Number of species</b>	<b>38</b>	<b>35</b>	<b>27</b>	<b>30</b>	<b>32</b>	<b>31</b>	<b>26</b>	<b>30</b>	<b>28</b>	<b>24</b>	<b>23</b>	<b>26</b>	<b>26</b>	<b>28</b>

After two years of organic (10-30 Mg ha<sup>-1</sup>) and mineral fertilization (30-50 kg N ha<sup>-1</sup>+30 kg P ha<sup>-1</sup>), changes were found in the botanical composition, with a diminution in the percentage of *Nardus stricta* by 3-8% and of *Festuca rubra* by 10-14%, and even of *Agrostis capillaris*, in variants with higher manure rates, by 5-14%, and an increase in the percentage of *Cynosurus cristatus* by 8-29% and of *Trifolium repens* by 1-5% (variants 4-7). The contribution of grasses to the composition of vegetation was 66-91%, but legumes were only of 2-11%, and species from other botanical families were 7-15%. On *Nardus stricta* grassland our determinations found 30 species were present, comprising 6 grasses, 2 legumes and 22 species from other botanical families, the dominant species being *Nardus stricta* (76%), followed by *Festuca rubra* (5%), *Agrostis capillaris* (3%) and *Anthoxanthum odoratum* (3%). After the two years of mineral fertilization, at rates of N<sub>100-200</sub>P<sub>100-200</sub>, the grassland botanical composition had changed significantly, the dominant species, *Nardus stricta*, diminishing its percentage by 44-67%, and its place being taken by *Festuca rubra* and *Agrostis capillaris*, at 26 and 25%, respectively, 21-47%, which became dominant species in the canopy (Table 1). On both the *Agrostis capillaris* - *Festuca rubra* grassland and the *Nardus stricta* grassland, the total number of species has decreased in fertilized variants, by 3-12 species in the case of organic and mineral fertilization and by 2-7 species in the case of mineral fertilization alone, irrespective of the type of fertilizer (Table 1).

## Conclusions

The management of permanent grasslands, in terms of usage, type and intensity of fertilization and method of control has a great influence on phytocoenotic biodiversity, on rate of species in the structure of vegetation, and dominant species in the canopy. Mineral fertilization with rates of 100 and 200 kg N ha<sup>-1</sup> + 100 and 200 kg P ha<sup>-1</sup> resulted in diminishing the percentage of *Nardus stricta* from 76% to 9-32% and increasing the percentage of the relatively valuable species of *Agrostis capillaris* and *Festuca rubra*, from 3% to 21-55%, and from 5% to 16-40%, respectively, but also in diminishing the total number of species from 30 to 23-28. The organic (10-30 Mg ha<sup>-1</sup>) and mineral fertilization (30 + 50 kg N ha<sup>-1</sup> + 30 kg P ha<sup>-1</sup>) resulted in changes in the botanical composition, by diminishing the percentage of *Nardus stricta* by 3-8% and of *Festuca rubra* by 10-14%, and even *Agrostis capillaris*, in higher manure rate variants, by 5-14%, and increasing the percentage of *Cynosurus cristatus* by 8-29% and *Trifolium repens* by 1-5%. The total number of species was diminished by 3-12. Most of the species which vanished from the vegetation canopy were forbs that were not considered of high fodder value.

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## **Session 1B**

### **Productivity in cultivated grasslands**

## Simulating the morphogenesis and cutting of perennial ryegrass (*Lolium perenne*) plants with a 3D model

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### Abstract

The canopy structure of grasslands is a major determinant of their use value, as it affects the quantity and the quality of the removed forage when mowed or grazed. The individual plant architecture determines the structure of the sward, and is highly sensitive to variations of the environment and to management practices. Several authors have suggested that this plasticity is partly mediated by a self-regulation process of the architecture. For instance, the height of the pseudostem could regulate the length of the leaves growing within it. To test this hypothesis we built a functional-structural 3D model of a ryegrass plant. The model uses the Lindenmayer-systems formalism (description of an object as a set of sub-units) and is based on the implementation of self-regulation rules. It satisfactorily captures the main static and dynamic architectural traits of ryegrass in non-limiting environmental conditions, so that a self-regulation process seems plausible. This model also allows testing the effects of cutting on the subsequent architecture development. This approach should ultimately allow us to better understand the morphogenetic variations and the evolution of the genotypic composition of simulated heterogeneous mono-species populations under changing environments.

Keywords: *Lolium perenne*, functional-structural modelling, ryegrass defoliation

### Introduction

We are lacking a proper understanding of what are the utmost determinants of the plasticity of the canopy structure in grasslands (e.g. trophic limitations, morphogenetic adaptations, etc.), because of the multiple interactions of the effects of environmental factors. The modelling approach allows us to study the separate effects of factors that are difficult to disconnect *in vivo*. More particularly, functional-structural plant models (Prusinkiewicz *et al.*, 1999), which explicitly describe the plant architecture using 3D computer graphics, are useful to analyse problems in which the spatial structure of the system is an essential property (Vos *et al.*, 2007). Our aim is to develop a functional-structural model of ryegrass morphogenesis, defoliation and regrowth. The plant model simulates the 3D development of the aerial parts of individual plants in the vegetative phase. A representation of the architecture of the grasses can be obtained by a description of the topology of the plant and of the leaf dynamics (e.g. growth kinetics, lifespan, geometry, etc.) at different levels of organization: leaf, tiller and whole plant. Basically, a realistic representation of the space occupation by the plant relies on a proper description of the number of leaves and tillers, and of the space orientation and shape of these phyto-elements. From a functional point of view, the processes controlling the triggering, coordination and kinetics of the leaves and tillers growth are of utmost importance. The main static (e.g. final leaf length) and dynamic (growth rate, elongation duration) characteristics of the leaves can be described as a function of their phytomer relative number (Evers *et al.*, 2005). On the other hand, several authors (e.g. Fournier *et al.*, 2007) suggested that some properties are controlled, in an indirect manner, by the pre-existing architecture. For example, the rapid growth phase of a given leaf could be triggered when the older leaf emerges out of the pseudostem cylinder. In our work, to test these hypotheses, we designed a

model that is as 'self-regulated' as possible, such that the only descriptive input is the general form of the growth function of the leaves (sigmoid).

## Materials and methods

According to the L-system approach the plant is modelled as a string of sub-units called modules. The development of the plant is simulated by rewriting the string according to procedural rules. This formalism allows the creation of realistic 3D representation by associating a volumetric object to each module. All these steps are performed in the LPFG simulation platform (Karkowski, 2002), on the basis of coded instructions. Here the structural representation of the plant is based on virtual phytomers, as described by Fournier *et al.* (2007), made of three types of modules standing for internodes, leaves and axillary buds (roots are not considered). Phytomers are emitted by modules representing shoot apical meristems. The functional part of the model relies on the emission of new phytomers and on the growth of leaves. Leaves grow according to a Beta-type function and their developments are coordinated by the detection of a specific event: the emergence of the leaves out of the tube made by the sheaths of the previous leaves. When this event occurs it activates the following developmental rules: i) the leaf determines the final length it will reach and its total leaf elongation duration (LED) according to the time elapsed since it has begun to grow, ii) a new phytomer is added and its own leaf starts its growth, iii) the axillary bud located two nodes below becomes a new tiller; it differentiates into a meristem, and emits its first phytomer which own leaf starts its growth. Regulation of tillering is achieved by decreasing the site filling to mimic the effects of self-shading. In this case we diminish the actual probability for a bud to differentiate as the plant total leaf area increases. In order to simulate the cutting, all the elements that are situated above the specified defoliation height are removed. The user parameterization of the model affects the relationship between the state of the leaf and the time it spent growing within the whorl. The user also parameterizes the surface thresholds for the tillering regulation, and the 3D representation (e.g. shape of the leaves, etc.). Using the methodology described above, the model automatically creates morphological patterns and is able to react to a cutting event (Figure 1). The morphogenesis is an emergent property resulting from the recursive development of the successive leaves, it is not deterministically dictated by the user.



Figure 1. Sequence of 5 images illustrating the growth, defoliation and regrowth of the 3D virtual plant.

## Results and discussion

On the basis of the few imposed developmental rules and subsequent parameterization, the model is able to satisfactorily capture the evolution of the patterns and kinetics of the major architectural traits (Figure 2). After a cutting event, the model response qualitatively mimics experimental observations. For instance, regrowth is characterized by shorter leaves and by an

increase in leaf and tiller appearance rates, due to the decrease of the pseudostem height and to the reduction of the total leaf area (inducing an increase of the site filling).

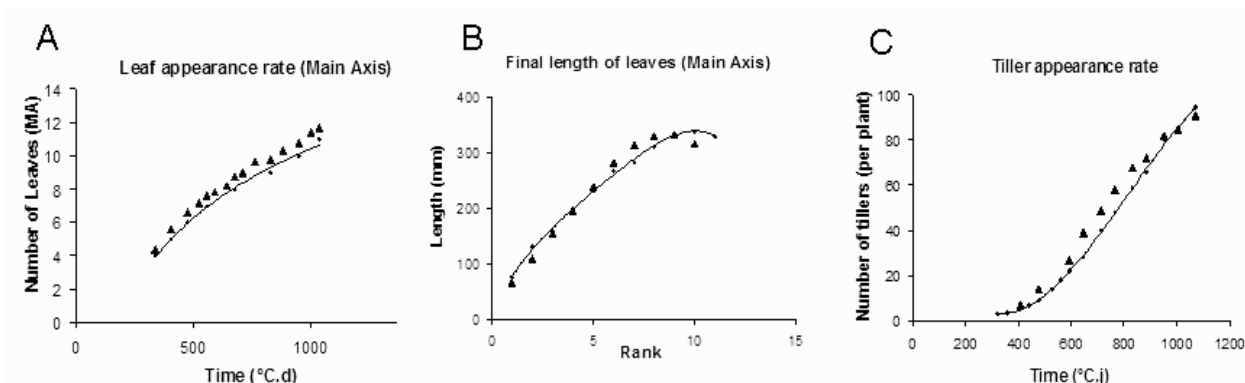


Figure 2. Simulated (▲) and observed (solid lines) Leaf Appearance Rate (A), Final length of leaves (B) and Tiller Appearance Rate (C). The correlation coefficient approximates 0.98 for these three traits. Data corresponding to the observations were collected on *Lolium perenne* plants, grown in growth chambers in 10x10 cm pots, 17 °C, 14 hours photoperiod, 500  $\mu\text{mol m}^{-2} \text{s}^{-1}$  P.A.R.

Finally, a self-organized system is able to partly simulate the plant development and regrowth, with no need to invoke other determinisms, so that the self-regulation of the architecture seems a plausible hypothesis. Even if some improvements of the model are needed, especially for the regrowth simulation, these results could lead us to rethink the relative importance of specific reactions (e.g. genetically determined) versus 'contingency' in the morphogenesis process.

## Conclusions

On the assumption that ryegrass architecture is a self-regulated process, the automated model presented here realistically mimics some of the main morphogenetic features of the plant before and after a cutting event. In the near future, this plant model will be used to simulate genotypically heterogeneous populations and to study how competition for the light resource affects the canopy structure and population composition under cutting constraints. This could open perspectives towards ecological studies.

## Acknowledgments

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# Sensor-based technologies to assess grassland biomass in short term leys

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## Abstract

In the past decades sensor-based technologies have gained increasing importance as part of precision farming applications. These methods have been developed particularly in arable crop production, while in grassland and forage production very little experience exists. A compilation of examples of sensor applications for the assessment of grassland yields is given with a special emphasis on the prediction of biomass by ultrasonic distance measurements. Results from a field experiment with pure swards and two-species mixtures composed of *Lolium perenne*, *Trifolium repens* and *Trifolium pratense* indicate linear relationships between sward height and biomass at common silage cutting dates within two different growth periods ( $R^2 = 0.88$  and  $0.78$  respectively). Improvements are possible regarding species composition. In swards with mature herbage predictions were less accurate and show a saturation effect for sward height while biomass still increases. It is suggested that ultrasonic sward height measurements provide a feasible tool to predict yields in species-poor grassland

Keywords: grassland, non-destructive, yield, spectrometry, ultrasonic, precision farming

## Introduction

The history of precision farming started in the late 1980s when the global positioning system (GPS) was introduced to agriculture. From the beginning, yield maps were of great interest (Blackmore, 2003). While the development of sensor-based technologies for this purpose proceeded rapidly for grain crops, these applications were rarely adopted on grassland. There is a considerable need for information about the variability of grassland sward characteristics in order to identify site-specific productivity levels, assess forage quality and optimize nutrient management. We focus in this study on a set of non-destructive survey approaches. Static measurements of sward height or tiller density to predict grassland biomass are well known, using methods such as the sward stick, rising plate meter, capacitance stick or sonic sward stick, and predominantly applied to grazed sward (Hutchings, 1991; Murphy *et al.*, 1995). However, only mobile geo-referenced measurements operate in a fast and efficient way to capture data with a high spatial resolution for the creation of maps, e.g. to express within-field heterogeneity. Ehlert *et al.* (2003) implemented an online mechanical sensor, which is now on the market, for recording the displacement of a pendulum related to plant mass. Other approaches are using contact-free methods like spectral-radiometric measurements which, in addition to biomass estimations (Biewer *et al.*, 2008), provide forage quality information (Schut *et al.*, 2003). Here, we present results on the use of an ultrasonic sensor to measure sward height and predict grassland yields. Most of the above-named methods have been developed in species-poor short-term grass leys, with the advantages of (a) an easy identification and verification regarding structural canopy effects, and (b) a high potential to be adopted in intensively managed grassland.

## Materials and methods

In 2006 a field experiment was carried out at the experimental site 'Hebenshausen' of the University of Kassel, on a plateau in the central hill region of Germany (51°23'N, 9°54'E,



240 m a.s.l.). The site was characterized by a loamy soil with pH 6.7, site-specific optimum contents of P, K and Mg, annual rainfall of 650 mm and an annual mean temperature of 9 °C. Pure swards and two-species mixtures (grass and legume) of perennial ryegrass (*Lolium perenne* cv. Fenema), white clover (*Trifolium repens* cv. Klondike) and red clover (*Trifolium pratense* cv. Pirat) were sown in autumn 2004 in plots of 3 x 10 m size. No fertilizers were used. During the first two growth periods in 2006, sward height was measured once a week using an ultrasonic sensor of type UC 2000-30GM-IUR2-V15 (Pepperl and Fuchs). The sensor was placed at a defined height in a framework above sampling plots of 50 x 50 cm. Measurements were replicated five times at each sample area with one position in the centre and four crosswise locations at a radius of 15 cm around the centre. Subsequently, samples were cut, oven-dried at 65 °C and dry matter yields calculated. The measurement periods were subdivided into three survey intervals: 3-24 May – at common silage cutting dates, 31 May-12 June – representing delayed cuts, and finally 3-24 July which was the second growth period. Statistical analyses were made using the general linear module in SAS 9.1 for regression analysis ( $P < 0.05$ ) including the treatment as a co-variable. The Tukey test was used for pairwise comparisons between treatments ( $P < 0.05$ ).

## Results and discussion

Grassland biomass could be predicted with linear regressions in the first interval, 3-24 May of growth period 1, as well as in growth period 2 (Table 1 and Figure 1). For older swards (31 May-12 June) predictions were less accurate and showed a quadratic relationship with a saturation effect caused predominantly by red clover. It depicts that with increasing biomass, sward height reacts less sensitively and finally descends, probably because of compaction. Also in the delayed cutting period pure white clover swards grew higher than those in younger growth stages at comparable yield levels, probably due to flower emergence. These aspects exhibit the limits in the development of a common equation for swards at different phenological stages. While in growth period 1, relationships were not significantly affected by sward composition, the covariate analyses indicated significant differences between treatments in growth period 2 (Table 1). Considering this fact, significant linear relationships could be found for pure swards of ryegrass ( $r^2 = 0.43$ ) and red clover ( $r^2 = 0.4$ ) as well as for mixtures of white clover / ryegrass ( $r^2 = 0.91$ ) and red clover / ryegrass ( $r^2 = 0.77$ ).

Table 1. Statistical summary of regression and covariance analyses (y = ultrasonic sward height in cm, x = dry matter yield in t ha<sup>-1</sup>. Treatments represent sward composition)

	1 <sup>st</sup> growth period		2 <sup>nd</sup> growth period
	3 <sup>rd</sup> May – 24 <sup>th</sup> May	31 <sup>st</sup> May – 12 <sup>th</sup> June	3 <sup>rd</sup> July – 24 <sup>th</sup> July
Function	$y = 0.2998x + 7.465$	$y = -0.33x^2 + 8.82x + 16.1$	$y = 9.41x + 10.09$
Coefficient of determination	$r^2 = 0.88$	$r^2 = 0.59$	$r^2 = 0.78$
Covariance of treatment	$p = 0.13$	$p = 0.41$	$p = <0.0001$

Ultrasonic reflections are affected by plant canopy structure (Hutchings, 1992). In this study coefficients of variance (CV) were very high in pure ryegrass (9.7-26.7%). White clover contributes to a smoother surface and showed lower CVs in pure swards (5.3-1.8%), but was overgrown by grass shoots in mixtures, resulting in higher CVs (11.3-14.5%). The significantly lowest CVs were detected in red clover pure swards (1.1-4.5%) and mixtures (1.8-7.3%), respectively. The variance of measurement replications was lower in older swards of growth period 1, which indicates that canopies appear to get more homogenous during growth development within the sensor surrounding. In the growth period 2, CVs were higher

than in the first, probably due to more individuals in generative state. Even if the canopy seems to be flat, in the case of red clover, the results showed that the accuracy of yield predictions depends additionally on the state of biomass development (Figure 1).

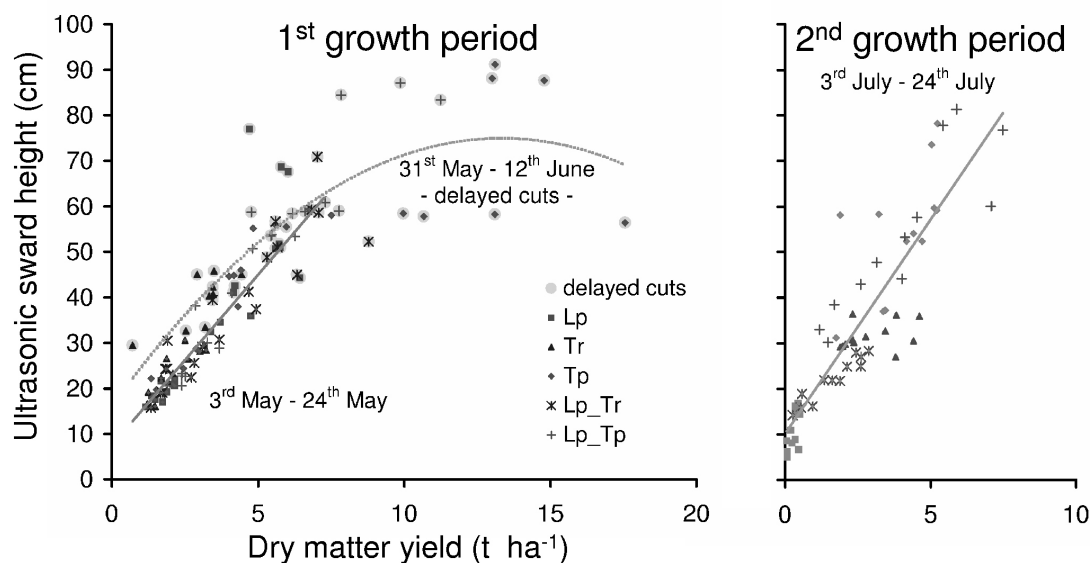


Figure 1. Relationships between ultrasonic sward height and dry matter yield of pure swards (*Lolium perenne* (Lp), *Trifolium repens* (Tr), *Trifolium pratense* (Tp)) and two-species mixtures (*L. perenne* with *T. repens* (Lp\_Tr) and *T. pratense* (Lp\_Tp)). The time periods indicate grouped calculations for the displayed regression functions.

## Conclusions

Although the mechanisms behind the relationship between sonic sward height and biomass remain unclear (Hutchings, 1992) the results suggest potential for this method for obtaining satisfactory accuracy in short-term leys with limited species diversity. Best results could be achieved at common silage cutting dates. Further improvements are possible for adjusting functions with respect to species composition.

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# Productivity effects of grass-legume mixtures on two soil types

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## Abstract

In 2003-2006, an experiment was carried out on mineral (Cambisol) and organic (Histosol) soil conditions to investigate the effects of different grass-legume mixtures on the DM yield and botanical composition of the swards. The experiment was established according the common protocol of COST Action 852, on 9 m<sup>2</sup> plots in a simplex design with 4 monocultures and 11 mixtures of the 4 species (fast establishing *Lolium perenne* and *Trifolium pratense* and slow establishing *Dactylis glomerata* and *Trifolium repens*), which were sown at two levels of overall sown abundance (low being 60% of high). Experimental communities were managed by cutting. Annual application of nitrogen was 90 to 120 kg ha<sup>-1</sup>. The dry matter (DM) yield and the proportion of sown and unsown species in the yield were determined. It was found that the above-ground biomass of four-species mixtures was significantly higher than that obtained from monocultures. The proportion of unsown species was higher in legume monocultures than in mixtures and grass monocultures. The soil types influenced slightly the botanical composition of the sward, and there was a much higher DM yield from the grass-legume mixtures on the mineral soil.

Keywords: grass-legume mixtures, botanical composition, yield, soil type

## Introduction

Legume-based systems are known to contribute towards sustainable, environmentally sensitive and energy efficient agriculture and are likely to assume an increasing importance (Tilman *et al.*, 1996). Environmental, biotic and management factors affect the productivity of the grass-legume mixtures (Peeters *et al.*, 2005). One of these is the soil type. The aim of this research was to study the effect of two soil type on DM yield and botanical composition of grass-legume mixtures.

## Materials and methods

The study was carried out during 2003-2006 at the Brody Experimental Station (52°26'N, 16°18'E) of the Agricultural University, Poznań, to evaluate the effect of different grass-legume mixtures on the DM yield and botanical composition of the sward in mineral soil (Cambisols; pH<sub>KCl</sub> – 5.9, N<sub>t</sub> – 1.1%, P<sub>2</sub>O<sub>5</sub> – 21.8 mg 100 g<sup>-1</sup>, K<sub>2</sub>O – 17.4 mg 100 g<sup>-1</sup>, Mg – 5.9 mg 100 g<sup>-1</sup>, altitude of 94.2 m a.s.l.) and organic soil (Histosols; pH<sub>KCl</sub> – 6.5, N<sub>t</sub> – 6.7%, P<sub>2</sub>O<sub>5</sub> – 17.5 mg 100 g<sup>-1</sup>, K<sub>2</sub>O – 30.0 mg 100 g<sup>-1</sup>, Mg – 7.1 mg 100 g<sup>-1</sup>, altitude of 91.4 m a.s.l.). The basic design with 30 plots according to the common protocol of COST Action 852 included 4 monocultures and 11 mixtures consisting of varying proportions of 4 species: fast establishing G1-*Lolium perenne* (cv. Lacerta) and L1-*Trifolium pratense* (cv. Merviot) and slow establishing G2-*Dactylis glomerata* (cv. Accord) and L2-*Trifolium repens* (cv. Milo). The 11 mixtures consisted of 4 mixtures dominated in turn by each species (70% of dominant and 10% of each of the other species), 6 mixtures dominated in turn by pairs of species (40% of each of two species and 10% of the other two) and the centroid community (25% of each species). The monocultures and mixtures were sown at two levels of overall sown abundance (low being 60% of the recommended: G1-47, G2-27, L1-22, L2-13 kg ha<sup>-1</sup>). The experiment was established on 9 m<sup>2</sup> (3 m × 3 m) plots in a simplex design (Ramseier *et al.*, 2005). Experi-

mental treatments were randomly assigned to plots, sown in autumn 2003 and managed by cutting. Fertilizer was applied each year at the following rates: 90-120 kg N ha<sup>-1</sup> (as 30 kg ha<sup>-1</sup> before each regrowth), 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, 90 kg K<sub>2</sub>O ha<sup>-1</sup>, and 3-4 regrowths were harvested. The yearly mean temperature and total precipitation for 2004, 2005 and 2006 was 9.1, 8.9, 9.6 °C and 584.7, 665.3, 549.4 mm, respectively. The DM yield and the proportion of sown and unsown species in the yield were determined. For estimation of yield (total annual above-ground biomass including unsown species) a subplot (6 m<sup>2</sup>) was cut to a height of 5 cm. Samples for botanical composition were taken from a fixed quadrat (1m × 1m) situated at the centre of each plot and were separated into five groups: the 4 sown species and unsown species. Statistical analysis of the total annual yield data was carried out by ANOVA. Two analyses were performed: (i) evenness gradient – this analysis is based on classifying the 15 mono/mixture plots into 3 groups: L – low (monocultures, 8 plots), M – medium (mixtures dominated by one species, 8 plots), H – high (all other mixtures, 14 plots); (ii) legume gradient – this analysis is based on classifying the 15 mono/mixture plots into 5 groups: Leg1 – monoculture legumes (L1 or L2, 100% L–4 plots), Leg2 – mixtures with high legume proportion (L1 or L2 70% and L1 and L2 40%, 80% L–6 plots), Leg3 – centroid and other mixtures with 50% L (L1 and L2 25% and L1 or L2 40% but not both, 50% L–10 plots), Leg4 – mixtures with low legume proportion (L1 and L2 10%, 20% L–6 plots), and Leg5 – monoculture grasses (G1 or G2, 0% L–4 plots).

## Results and discussion

The soil type influenced the productivity of grass-legume mixtures. The higher DM yield was obtained on the mineral soil. The data in Table 1 show consistent positive effects of increasing plant evenness. In our study, the yield of four-species mixtures exceeded that expected from monoculture performances either on mineral or organic soils ( $P < 0.001$ ). This effect has been reported by many other authors, e.g. Tilman *et al.* (1996) and Peeters *et al.* (2005). There was no effect of density at either soil, nor was there an evenness × density interaction.

Table 1. Total annual DM yield (kg ha<sup>-1</sup>) at three levels of evenness for two soil types (averaged over years 2004-2006).

Evenness	Mineral				Organic			
	L	M	H	Mixed	L	M	H	Mixed
Mean	6877	8491	8404	8448	5579	6682	6910	6796
Values for comparing means of evenness	L vs. M	L vs. H	M vs. H	L vs. Mixed	L vs. M	L vs. H	M vs. H	L vs. Mixed
SED	475	421	421	381	394	349	349	316
t value	3.40	3.63	0.21	4.12	2.80	3.81	0.65	3.85
P	**	**	ns	***	**	***	ns	***

ns – non significant, \* $P < 0.05$ , \*\* $P < 0.01$ , \*\*\* $P < 0.001$ .

Total yield in grass-legume mixtures exceeded the yield of legume monocultures on both soil types ( $P < 0.001$ ). Yield of grass monocultures was not significantly different from mixture yields at either soil (Table 2). There were some monocultures, particularly G2 in second and third year of utilization, which yielded as much or more than mixtures. Additionally, some of the mixtures did not have very much legume in the sward, which essentially made them near-grass monocultures. There was also no effect of legume gradient × density interaction.

Table 2. Total annual DM yield (kg ha<sup>-1</sup>) at five levels of legumes in sown mixtures for two soil types (averaged over years 2004-2006).

Legume gradient	Mineral					Organic				
	Leg1	Leg2	Leg3	Leg4	Leg5	Leg1	Leg2	Leg3	Leg4	Leg5
Legume %	100	80	50	20	0	100	80	50	20	0
Mean	5558	8484	8451	8363	8197	4747	6588	6849	7029	6412
Values for comparing means of legume gradient	1 vs. 5	1 vs. 3	1 vs. 2	3 vs. 5	4 vs. 5	1 vs. 5	1vs. 3	1vs. 2	3vs. 5	4vs. 5
SED	437	366	399	366	399	459	384	419	384	419
t value	6.04	7.91	7.34	0.70	0.42	3.63	5.47	4.39	1.14	1.47
P	***	***	***	ns	ns	**	***	***	ns	ns

ns – non significant, \* $P < 0.05$ , \*\* $P < 0.01$ , \*\*\* $P < 0.001$ .

The soil types influenced slightly the botanical composition of the grass-legume mixtures and invasion of unsown species (Figure 1). The legume species disappeared from the sward during the years of utilization. The proportion of unsown species was higher in legume monocultures than in mixtures and grass monocultures.

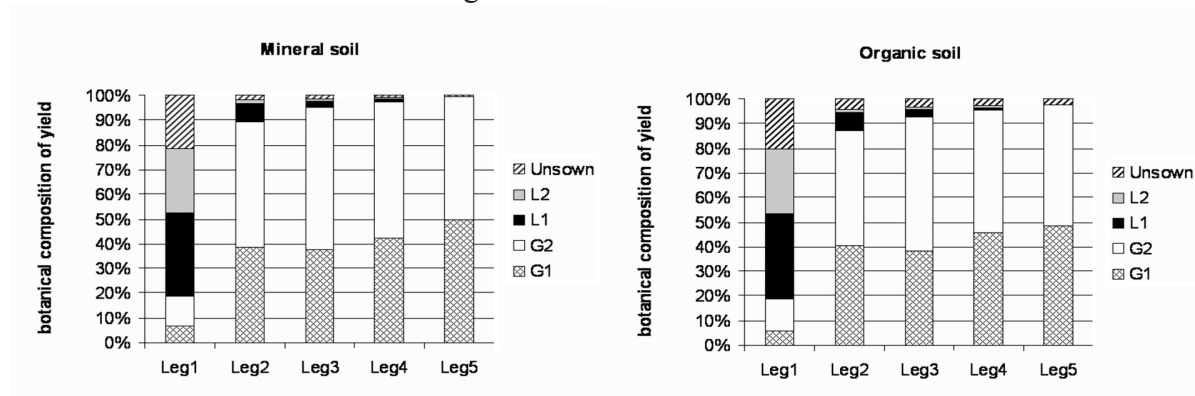


Figure 1. Botanical composition of the sward at five levels of legumes in sown mixtures for two soil types (averaged over utilization years 2004-2006).

## Conclusions

Productivity of four-species mixtures was significantly higher than that obtained from monocultures on both mineral and organic soils. The total DM yield in grass-legume mixtures exceeded the yield of legume monocultures on both soil types. Taking into consideration the legume gradient, the yield of grass monocultures was not significantly different from the yields of mixtures on either soil. A greater DM yield of grass-legume mixtures was obtained on the mineral soil. The soil types influenced slightly the botanical composition of the sward.

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# Well-balanced grass-legume mixtures with low nitrogen fertilization can be as productive as highly fertilized grass monocultures

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## Abstract

A pan-European agrobiodiversity grassland experiment showed that mixed swards with four grass and legume species had a greater yield than that expected from monocultures. We tested (i) at which legume proportion the highest mixture yield was achieved, (ii) whether overyielding still occurred in highly N-fertilized systems, and (iii) whether mixtures with low N fertilization produced yields comparable to grass monocultures with very high N fertilization. The effects of diversity on productivity under the influence of N fertilization (50, 150 or 450 kg N ha<sup>-1</sup> yr<sup>-1</sup>) was investigated in pure stands and mixtures with highly varying proportions of *Lolium perenne*, *Dactylis glomerata*, *Trifolium pratense* and *Trifolium repens*. To achieve a maximal yield over the three years, the sowing proportion of legumes should have been 63%, 56% and 46% at low, moderate and very high N, respectively. Though the extent of overyielding lessened with increasing N fertilization, it was still 47% at the very high N level. The main finding of the study was that overyielding was so strong that well-balanced mixtures with low N produced a similar yield to the highest yielding grass monoculture with very high N fertilization. We conclude that high productivity can be maintained and large quantities of N fertilizer be saved by the use of well-balanced grass-legume mixtures.

Keywords: fertilizer efficiency, symbiotic N<sub>2</sub> fixation, transgressive overyielding

## Introduction

Pure grass swards produce high yields provided there is a high input of mineral nitrogen (N) fertilizer. The application of large quantities of N fertilizer is expensive and its production very energy intensive and a source of the greenhouse gas CO<sub>2</sub>, which is why alternative systems with reduced N fertilizer input are necessary. In ecological studies carried out under nutrient-poor conditions, diverse plant communities are often more productive than those of low diversity (Hooper *et al.*, 2005). However, whether this also holds true for fertile agronomic conditions is unclear. Recently, a multi-site grassland experiment with regard to agricultural conditions showed that mixed swards of four species had a greater yield than monocultures under a large range of climatic conditions (Kirwan *et al.*, 2007; Helgadottir *et al.*, 2008). While studies on the effects of diversity mainly concentrated on the influence of species richness (number of species in the mixture), Kirwan *et al.* (2007) demonstrated that species evenness (the relative proportion of species in the mixture) is a major determinant for these diversity effects. The aims of our study, carried out under fertile agronomic conditions, were (i) to determine at which legume proportion mixture yield is maximized, (ii) to test whether overyielding still occurs at very high N fertilizer input and (iii) to ascertain whether diversity effects are so large that mixtures at low N can produce comparable yields to highly fertilized grass monocultures.

## Material and methods

Pure and mixed stands with widely varying proportions of the two grass species *Lolium perenne* L. and *Dactylis glomerata* L., and the two legume species *Trifolium pratense* L. and *Trifolium repens* L. were established following the simplex design (Cornell, 2002; for details see Helgadottir *et al.*, 2008). Swards were fertilized with 50, 150 or 450 kg N ha<sup>-1</sup> yr<sup>-1</sup>; hereafter referred to as low, moderate and very high N fertilization. Phosphorus and potassium were applied as recommended for intensively managed grasslands under the given growing conditions (40 kg P and 220 kg K ha<sup>-1</sup> yr<sup>-1</sup>). Since mixtures were sown with widely varying species proportions (from 10 to 70% for each individual species), the design allowed the estimation of species proportions in the seed mixture for maximal yield based on regression models (Kirwan *et al.*, 2007). The swards were harvested five times annually for three years after establishment. Total yield, N yield and N from symbiotic N<sub>2</sub> fixation (using the <sup>15</sup>N dilution technique) were determined.

## Results and discussion

The magnitude of overyielding (Trenbath, 1974) of centroid mixtures (equal proportions of all species) was 62%, 55% and 47% at low, moderate and very high N respectively, compared to the yield expected from the average yield of the four monocultures (Table 1). This strong advantage of the mixtures is surprising as all four species are high yielding and often cultivated in intensively managed fertile grasslands, with high yields of monocultures (Figure 1).

Table 1. Yield expected from the monocultures yield, effectively achieved yield, proportion in harvested N from symbiosis (N<sub>sym</sub>), and fertilizer N efficiency of the mixtures sown with equal proportions of *L. perenne*, *D. glomerata*, *T. pratense* and *T. repens* (centroid mixtures), as well as predicted yield of the optimal mixtures and the corresponding legume proportion for maximal yield for the three N fertilization levels. Estimations are based on regression models fitted to the observed data.  $DM_{yield}/N_{fert}$  = kg dry matter harvested per kg N applied.

	Centroid mixtures				Optimal mixtures	
	Yield expected from monocultures	Achieved yield	N <sub>sym</sub>	DM <sub>yield</sub> /N <sub>fert</sub>	Estimated yield	Legume proportion at sowing
	t DM ha <sup>-1</sup> yr <sup>-1</sup>	t DM ha <sup>-1</sup> yr <sup>-1</sup>	%	kg DM kg <sup>-1</sup> N	t DM ha <sup>-1</sup> yr <sup>-1</sup>	%
Low N	9.1	14.8	54	261.2	15.2	63
Moderate N	10.3	16.0	36	106.7	16.1	56
Very high N	12.0	17.7	14	41.2	17.9	46
SE	0.38	0.43	-	-	0.38	-

The magnitude of overyielding was slightly smaller than the 70% reported by Cardinale *et al.* (2007) from a meta-analysis of ecological studies. We suggest that the slightly greater effect they found was due to the fact that low yielding species were often included in the reviewed studies. At all three levels of N fertilization the yield of all the mixtures even exceeded that of the most productive monoculture (transgressive overyielding, Trenbath (1974)) (Figure 1). This occurred though mixtures were sown with widely varying species proportions (from 10 to 70% for each species). Such a clear and stable transgressive overyielding is an unexpected result since Cardinale *et al.* (2007) found that transgressive overyielding only occurred in 12% of the studies summarized in their meta-analysis. Nevertheless, the diversity effect on productivity was greater at high than at low species evenness, which is in agreement with the findings of Kirwan *et al.* (2007). To achieve maximal yield over the three years, the legume proportion at sowing should have been 63%, 56% and 46% at low, moderate and very high N respectively (Table 1). The response of the pure grass swards to N fertilizer was much greater

than that of the mixtures. Nevertheless, the yield of the grass monocultures fertilized at very high N did not exceed the yield of the highest yielding mixtures fertilized at low N (Figure 1). This is a key finding in our study, demonstrating the high forage production potential of grass-legume mixtures at low N fertilization. Symbiotic  $N_2$  fixation in centroid mixtures contributed 54%, 36% and 14% to the quantities of harvested N at low, moderate and very high N respectively (Tab. 1). The efficiency of fertilizer N (kg dry matter harvested per kg N applied) was correspondingly much greater at low than at very high N. The reduction of the quantities of fixed N with an increased N fertilization was due to both the down-regulation of the N fixing activity of legumes with the increased N availability (Nyfeler *et al.*, 2006) and a significant reduction of the legume proportion in the sward.

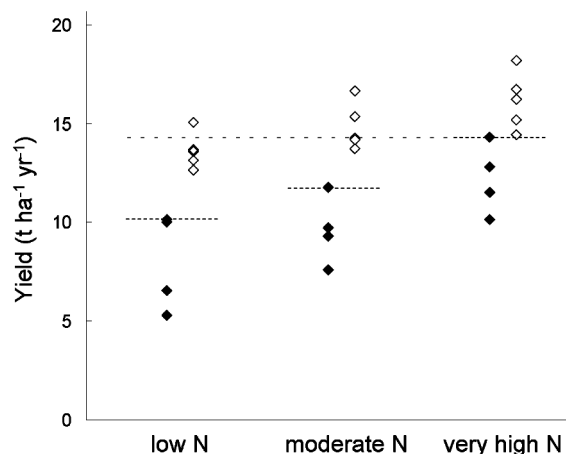


Figure 1. Dry matter yield of monocultures (♦) and mixtures (◇) for the three N fertilization levels. Transgressive overyielding is indicated by the dotted lines.

## Conclusions

Well balanced grass-legume mixtures fertilized with only  $50 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  produced a similar forage yield as the highest yielding grass monoculture at  $450 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ . This demonstrates that high productivity can be maintained while saving large amounts of N fertilizer in intensive systems by the use of grass-legume mixtures.

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## Herbs in grasslands – effect of slurry and grazing/cutting on species composition and nutritive value

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### Abstract

Herbs are established in many organic grasslands due to their expected beneficial properties for nutritive value and biodiversity. However, knowledge about grassland herbs is limited. Three mixtures were therefore established at different grazing/cutting management and slurry applications. The competitiveness of the species varied greatly. Plantain (*Plantago lanceolata*) and lucerne (*Medicago sativa*) competed best under cutting; in contrast, chicory (*Cichorium intybus*) competed best under grazing. Caraway (*Carum carvi*), burnet (*Poterium sanguisorba*) and lotus (*Lotus corniculatus*) had a relative low competitiveness. Slurry application did not affect the proportion of non-leguminous herbs. The nutritive value differed: caraway had the highest and plantain the lowest values, as judged by *in-vitro* organic matter digestibility (IVOMD) and content of NDF, ADF and ADL. Under cutting, the annual dry matter yields of the three mixtures were similar.

Keywords: herbs, competitiveness, nutritive value, grazing, cutting, slurry

### Introduction

On Danish organic dairy farms, herbs are often sown together with grass/clover – broadcast or strip sown. There is a considerable anecdotal support for the beneficial properties of herbs, but the amount of herbs in the sward is often limited (Smidt and Brimer, 2005). The expectations are that herbs contribute to greater biodiversity, better drought tolerance, higher N utilization in the cow, higher mineral content, reduced parasitism, and a positive effect on milk quality. However, available knowledge about herbs is very limited concerning their establishment, growth, nutritive value and different side effects. An experiment was therefore established to examine the effect of management on the competitiveness and the nutritive value of different species.

### Materials and methods

The study was carried out in an organic dairy cattle crop-rotation system running since 1987 at Research Centre Foulum. Three seed mixtures were established in 2006, and were composed of: 1) perennial ryegrass (*Lolium perenne*), white clover (*Trifolium repens*) and red clover (*Trifolium pratense*); 2) mix 1 together with the herbs chicory (*Cichorium intybus*), plantain (*Plantago lanceolata*), caraway (*Carum carvi*), lotus/birds foot trefoil (*Lotus corniculatus*), salad burnet (*Poterium sanguisorba*), sainfoin (*Onobrychis viciifolia*) and chervil (*Anthriscus cerefolium*); and 3) mix 2 together with festulolium (*XFestulolium*) and lucerne (*Medicago sativa*). The seeds were sown at 0-1 cm depth and the total rate was 26 kg ha<sup>-1</sup>. The seed rate of the single species is shown in Table 1. In 2007 the sward was either grazed continuously by heifers or cut four times. In addition, there were two application levels of cattle slurry: 0 and 200 kg total N ha<sup>-1</sup> (half in spring and half after the first cut). There were two replicates. During the second regrowth, parts of the grazing plots were fenced off and harvested at the same time as the cutting plots. Degree of coverage was estimated in 0.5 m<sup>2</sup> randomly positioned plots in early growth in April. The botanical composition at

harvest was determined by hand separation of sub samples. Herbage quality at cut 1 and 3 was determined for the single species; IVOMD (Tilley and Terry) and fibres (NDF, ADF and ADL; van Soest). Results from the first year, 2007, are reported in this paper.

## Results and discussion

Mix 1 represents a common mixture for pasture, with a small amount of red clover added (Table 1). In mix 2 herbs replace a part of mix 1 (5 out of 26 kg). Mix 3 has the highest species number and it is supplemented with lucerne and festulolium as longer lasting species (Table 1). The establishment of chervil and sainfoin did not succeed at all. The other herbs constituted 3 to 20% of the area in spring (Table 1). White clover was not established satisfactorily and constituted only 20% of the area in mix 1, which probably is a main reason for the fact that nearly half of the area was covered by herbs in mix 2 and 3. In the multi-species mixtures 2 and 3, the grass only constituted approximately 20% of the area.

Table 1. Seed rate of the different species, degree of coverage in April and botanical composition in mixture 3 in unfertilized cut plots during the season.

Mixture	Seed rate			Degree of coverage			Cut 1	Cut 2	Cut 3	Cut 4
	1	2	3	1	2	3	3	3	3	3
	% of total weight			% of area			% of herbage DM			
Festulolium			31			5.4				
Per. Ryegrass	82	66	28	61.0	21.8	16.8	23.3*	13.6*	4.0*	7.9*
White clover	14	12	5	19.9	10.1	8.4	2.6	7.9	14.3	16.0
Red clover	4	3	1	15.0	11.5	5.1	6.5	13.1	21.8	10.6
Chicory		3	3		16.1	15.0	10.9	9.0	5.8	3.7
Plantain		3	3		19.7	18.5	40.9	32.5	21.7	22.7
Caraway		3	3		5.3	6.7	2.1	0.5	2.1	1.5
Burnet		3	3		3.0	4.0	0.7	1.0	0.4	0.3
Lotus		2	2		3.3	3.1	0.8	2.9	4.9	2.9
Chervil		2	2		0	0	0	0	0	0
Sainfoin		3	3		0	0	0	0	0	0
Lucerne			15			8.9	10.7	19.1	25.1	33.7
Unsown sp.				2.4	1.8	1.8	1.5	0.5	0.5	0.1
Bare soil				1.7	7.4	6.7				

\*include both festulolium and perennial ryegrass.

The mean proportion of herbs was about 40% of the dry matter (DM) and was thus considerably higher than in multi-species pastures reported by Goh and Bruce (2005). Slurry application decreased the proportion of the legume species (white clover, red clover, lotus and lucerne), increased the grass proportion, but hardly affected the non-leguminous herbs (data not shown). During the season the proportion of legumes increased from 21 to 66% of DM ( $P = 0.002$ ) in unfertilized cut plots (Table 1). In the fertilized plots the legume proportion also increased considerably, from 14 to 54% ( $P = 0.03$ ). This was mainly at the expense of grass and plantain (Table 1). Grazing/cutting affected the botanical composition considerably (Table 2). In the grazed plots compared with the cut plots, the proportions of grass and white clover were higher and the proportions of red clover and lucerne were lower. This effect is normally found in traditional pastures, and was thus the same for multi-species mixtures. In contrast to chicory, the plantain proportion was reduced by grazing (Table 2). Thus, these two dominating herbs seem adapted to different growing conditions. Inclusion of lucerne and festulolium in the sward (mix 3 compared with mix 2) mainly depressed the content of red clover, whereas the herbs were nearly unaffected (Table 2). Even though the species

composition was very different between the mixtures, the annual yield under cutting was not affected by the mixtures (Table 3). Slurry application increased the yield from 12.2 to 14.1 t DM ha<sup>-1</sup> ( $P < 0.001$ ).

Table 2. Botanical composition in the second regrowth in cut and grazed plots, and herbage quality expressed as mean of first and third cuts across slurry and defoliation systems.

	Grazing			Cutting			Herbage quality*			
	Mix 1	Mix 2	Mix 3	Mix 1	Mix 2	Mix 3	IVOMD	NDF	ADF	ADL
Grass	23.1	15.4	19.1	19.8	6.2	8.8	77.8 <sup>b</sup>	47.9 <sup>a</sup>	28.7 <sup>b</sup>	2.4 <sup>e</sup>
White clover	34.6	18.6	14.2	27.6	13.6	11.8	77.7 <sup>b</sup>	26.6 <sup>e</sup>	23.3 <sup>d</sup>	4.0 <sup>b</sup>
Red clover	41.4	31.9	25.7	52.2	49.0	21.0	74.7 <sup>bc</sup>	30.6 <sup>d</sup>	23.2 <sup>d</sup>	3.2 <sup>cd</sup>
Chicory		18.1	15.8		6.4	7.5	73.2 <sup>c</sup>	32.6 <sup>d</sup>	27.6 <sup>bc</sup>	3.6 <sup>bc</sup>
Plantain		12.0	13.8		20.5	21.2	63.0 <sup>e</sup>	42.6 <sup>b</sup>	33.6 <sup>a</sup>	6.1 <sup>a</sup>
Caraway		2.0	1.1		1.7	1.7	82.4 <sup>a</sup>	25.8 <sup>e</sup>	23.5 <sup>d</sup>	2.8 <sup>de</sup>
Burnet		0.3	0.3		0.3	0.4				
Lotus		1.5	2.5		2.4	3.3	68.1 <sup>d</sup>	30.8 <sup>d</sup>	26.2 <sup>c</sup>	5.8 <sup>a</sup>
Lucerne			7.5			23.9	67.8 <sup>d</sup>	36.0 <sup>c</sup>	28.7 <sup>b</sup>	5.6 <sup>a</sup>
Unsown spp.	0.9	0.2	0.2	0.8	0.1	0.5				

\*different letters indicate significant differences ( $P < 0.05$ ) between species.

There was a great difference in nutritive value between the species (Table 2). Plantain differed from the others by having the lowest value: very low IVOMD and very high content of NDF, ADF and ADL. The opposite was shown for caraway, which differed by having the highest nutritive value. Chicory had roughly the same nutritive value as red clover according to the measured parameters. Lotus and lucerne were characterized by having the same low IVOMD and high ADL, whereas lotus had a lower NDF and ADF than lucerne. Sanderson *et al.* (2003) reported nearly the same nutritive value of plantain and chicory in monoculture, and this was not confirmed by the results shown here. In this multi-species experiment, however, the herbage quality is probably affected by the growing conditions for the individual species.

Table 3. Annual yield under cutting (t DM ha<sup>-1</sup>).

Mixture	1	2	3	1	2	3
Slurry app	0 N	0 N	0 N	200 N	200 N	200 N
t DM ha <sup>-1</sup>	12.7	11.9	12.1	13.8	14.3	14.2

## Conclusions

The competitiveness of the individual species in multi-species grassland was affected by species composition and management. The nutritive value differed very much. Plantain had the lowest and caraway the highest nutritive value.

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# Overwintering of timothy and perennial ryegrass in Norway from a climate change perspective

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## Abstract

In a comprehensive research programme, WINSUR, we are currently investigating the effect of climate change on overwintering of *Phleum pratense* L. and *Lolium perenne* L., two important forage grasses. We summarize results from different studies in the field and controlled environments. Increased CO<sub>2</sub> concentration in the atmosphere (+170 ppm) had no effect on frost hardening in an open-top chamber experiment, whereas a temperature increase of 2 °C in the autumn reduced the frost tolerance by 1 °C. Plants transferred from the field to a phytotron in January dehardened rapidly. The most winter hardy cultivar dehardened from -26 to -18 °C during one week of exposure to 3 °C, and to -15 °C during one week exposure to 9 °C. The data suggest that winter-hardy cultivars of northern origin may be as prone to dehardening as less winter-hardy cultivars of southern origin. Ice encasement and frost tolerance were highly correlated. The results are discussed from a climate change perspective.

Keywords: forage grasses, climate change, winter survival, ice encasement, frost, CO<sub>2</sub>

## Introduction

Climate change scenarios for Norway (RegClim, 2005; <http://regclim.met.no>) indicate that the temperature will increase, especially in the winter (up to 3.5 °C) and in the northern parts of the country. There will be more frequent events of extreme precipitation, especially along the coast, more precipitation as rain and less as snow, and more variable weather in general, although the extent of variation is under debate. What will be the yields and winter persistency of swards with perennial grasses in the future climate? In a comprehensive research programme in Bioforsk, WINSUR, we are currently investigating the winter persistency of timothy (*Phleum pratense* L.) and perennial ryegrass (*Lolium perenne* L.), two important forage grasses with different growth strategies and winter hardiness, in the predicted future climate. We summarize results from different experiments carried out in the field and controlled environments.

## Materials and methods

Two contrasting cultivars of timothy and two of perennial ryegrass were compared with respect to growth and development of frost and ice encasement tolerance. The experimental sites were Holt (69.65°N, 18.91°E), Kvithamar (63.49°N, 10.88°E), Fureneset (61.29°N, 5.04°E) and Særheim (58.76°N, 5.65°E). The effect of increased autumn temperature (*ca.* 2 °C above the ambient temperature) and CO<sub>2</sub> concentration (*ca.* +170 ppm) on growth and hardening was investigated in open top chambers at Særheim in 2005/2006 using pot grown plants. The ambient daily mean temperature was 8 °C for the period 1 September to 15 December. The development of frost tolerance was studied at Holt and Særheim in 2005/2006

using pot grown plants with the pots placed close to each other in the field to mimic normal sward conditions. There were 10 plants per pot and 20 pots per m<sup>2</sup>. Tillers were sampled at various points in time to determine their frost tolerance which was expressed as LT50 (the temperature below which at least 50% of the population was killed) (Larsen, 1978). The simultaneous development of frost and ice encasement tolerance was studied at Holt, Kvithamar and Særheim in 2006/2007 in field plots. To facilitate sampling in periods with frozen soil, turfs for later sampling were dug up in the end of autumn and placed in growth containers (30 cm x 40 cm x 17 cm) adjacent to the field (Holt) or placed back in their original position in the field with a 3 mm felt underneath (Kvithamar). At Særheim, all sampling could be done directly in the field due to limited soil frost. Tillers were sampled to determine their frost tolerance (as described above) together with their ice encasement tolerance which was expressed as LD50 (the number of days required to kill 50% of the population when the tillers were completely encapsulated in ice and stored in darkness at -2 °C) (Gudleifsson and Björnsson, 1989). A dehardening experiment was carried out at Holt in 2006. Plants grown outdoors in pots were brought indoors in January, tested for LT50 and grown in a phytotrone at different temperatures for subsequent LT50 tests. The day length was 12 h. All the LT50 and LD50 values presented were estimated using probit analysis.

## Results and discussion

Increased CO<sub>2</sub> concentration stimulated the accumulation of biomass in autumn (total dry weight above ground 18.5 and 22.6 g pot<sup>-1</sup> at ambient and elevated CO<sub>2</sub>, respectively, on 21 November;  $P < 0.05$ ) but had no effect on the frost tolerance (average LT50 -12.2 °C on 15 December) or the density of tillers in spring (average 123 pot<sup>-1</sup> on 6 April). There was no interaction between CO<sub>2</sub> and temperature for any parameter. In a corresponding experiment carried out at the same location in a previous year, there was similarly no effect of CO<sub>2</sub> on the frost tolerance of winter wheat (Mortensen and Hanslin, 2007).

Increased autumn temperature, on the other hand, delayed hardening and reduced the frost tolerance. This was evident both in the open top chamber experiment at Særheim, although the effect was modest (LT50 on 15 Dec. -12.7 and -11.7 °C at ambient and high temperature, respectively,  $P < 0.05$ ) and when comparing the development of frost tolerance in plants grown outdoors under contrasting weather conditions (Table 1).

Table 1. Frost tolerance (LT50; °C) in timothy and perennial ryegrass at the end of November/beginning of December, together with the autumn temperature (daily mean air temperature in the previous month, °C). Averages for two cultivars of each species are given. 95% Fiducial limits for LT50 were within  $\pm 1$  °C except for the ryegrass at Holt in 2005 ( $\pm 2$  °C).

Year	Holt			Særheim		
	Autumn temp.	LT50 timothy	LT50 ryegrass	Autumn temp.	LT50 timothy	LT50 ryegrass
2005	5.6	-17.4	-10.8	5.8	-19.9	-12.2
2006	2.3	< -20	< -17	7.4	-17.6	-9.8

Thus, unusually mild autumn conditions led to poorer frost tolerance at Holt in 2005 than in 2006 when the conditions were more normal (colder). Similarly, at Særheim the poorest frost tolerance was obtained after the warmest autumn (2006). At the same time, it is evident that other factors also play a role in the development of frost tolerance. Differences in day length and radiation during the autumn most likely contributed to the differing results between Holt and Særheim. The presence or absence of snow modifying the conditions around the apex is also an important factor for frost hardening. Mild weather in mid-winter may melt the snow and promote dehardening. Plants at Holt transferred from the field to a phytotrone with temperatures between 3 and 9 °C in January 2007 dehardened rapidly (Table 2).



Table 2. Frost tolerance (LT50, °C) in two timothy cultivars in the field and one week after they had been transfer to a phytotron and subjected to three different growing temperatures. The upper and lower 95% Feducial limits of the LT50 values are presented in brackets.

	In the field	One week after transfer to phytotron at		
		+3 °C	+6 °C	+9 °C
Engmo	-26.0 (-23.9/-30.4)	-17.6 (-16.9/-18.3)	-15.6 (-14.9/-16.4)	-14.7 (-14.0/-15.4)
Grindstad	-15.5 (-13.6/-16.7)	-14.2 (-13.2/-15.6)	-12.0 (-11.2/-12.8)	-8.6 (-7.4/-9.6)

Substantial dehardening was observed even at the lowest growing temperature 3 °C. Both ryegrass cultivars similarly dehardened rapidly (data not shown). The results confirm earlier findings (Jørgensen *et al.*, 2006) and suggest that rapid hardening may occur in periods of mild weather, even in mid-winter, and that northern, winter-hardy cultivars like Engmo may be as prone to dehardening as less winter-hardy cultivars like Grindstad.

There was a large genetic variation in frost tolerance. The lowest LT50 value observed in the different experiments together was -27, -23, -18, and was -16 °C for Engmo, Grindstad, Riikka and Gunne, respectively. There was similarly a large variation in ice encasement tolerance, with Engmo reaching a maximum LD50 greater than 9 weeks, followed by 7, 3 and 2 weeks for Grindstad, Riikka, and Gunne, respectively. Furthermore, LD50 varied between locations and time of season (Table 3), generally with the highest values at Holt and the lowest at Særheim. The LD50 values were highly correlated with the LT50 values measured at the onset of ice encasement. Since the LT50 can be modelled with good precision, the close correlation may facilitate modelling of LD50 and ice encasement injury.

Table 3. Ice encasement tolerance (LD50; days) in Grindstad timothy and Gunne perennial ryegrass at three different locations in 2006/2007. 95% Feducial limits for LD50 were within  $\pm 2$  days for ryegrass and  $\pm 3$  days for timothy except for at Særheim in March ( $\pm 8$  days).

Location	Timothy			Ryegrass		
	December	January	March	December	January	March
Holt	> 44	46	32	14	15	11
Kvithamar	41	41	29	9	> 12	8
Særheim	35	30	30	10	< 4	n.a.

## Conclusions

The predicted temperature increase in autumn temperatures will delay the hardening and reduce the frost tolerance of the genotypes presently grown. Increased CO<sub>2</sub> concentration in the atmosphere will have limited effect on frost hardening. Rapid dehardening may occur as a result of mild weather in mid-winter, even in winter-hardy cultivars of northern origin. Modelling work is in progress that will give more detailed estimates of how timothy and perennial ryegrass will perform in the future winter climate.

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# **Yield potential, correlation and path-coefficient for two synthetics and three commercial varieties of alfalfa**

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## **Abstract**

During 2003-2005, a field trial was conducted at the New Valley Research Station to evaluate the yield potential, correlation and path-coefficient of two synthetics of alfalfa (Syn<sub>1</sub> and Syn<sub>2</sub>; second and the third combination generations, respectively) compared with three commercial varieties (Ismailia-1, Ismailia-94 and Siwa). Syn<sub>2</sub> had significantly higher fresh-weight and dry matter forage yields (171.1 and 46.9 t ha<sup>-1</sup>, respectively) than other entries and ranked first for all traits. Significantly positive correlations were found among the studied traits. The path-coefficient analysis indicated that the number of tillers was the most prominent direct effect upon fresh forage yield (51.82%) with relative importance of 25.9%, followed by plant height (42.41%) with relative importance of 21.3%.

Keywords: Alfalfa, synthetics, correlation, path coefficient.

## **Introduction**

In Egypt there is a gap between the demand and the consumption of green forages. This is especially the case during the summer where the available forages are limited due to competition from strategic crops cultivated on the limited arable land. Alfalfa is the best crop to overcome this problem as it is the most suitable forage crop to be cultivated on newly reclaimed land. It is also an important key to the amount and quality of forage production and longevity of the stand. The correlation studies give the amount of association between any pair of characters. The direct and indirect effects of each of the components are not revealed by these correlation studies especially when more variables are included. Hence, path-coefficient is preferable than correlation coefficient as the path-coefficient measures the direct and indirect effects of characters influencing the yield. Therefore, this investigation was conducted to evaluate two new developed alfalfa synthetics compared with three cultivars and to measure the direct and indirect effects and the relative importance of the characteristics influencing the yield potential of alfalfa.

## **Materials and methods**

Three local cultivars of alfalfa (*Medicago sativa* L.); Ismailia-1, Ismailia-94 and Siwa, and two synthetic populations developed at Giza Research Station (Syn<sub>1</sub> and Syn<sub>2</sub>) were used in this investigation, conducted at the New Valley Research Station during 2003-2005. A randomized complete block design with three replicates was used. The plot size was 4m<sup>2</sup> and the seed was drilled on 1 September 2003 in 6 rows, 20-cm apart, at the rate of 47.6 kg ha<sup>-1</sup>. All the cultural practices were applied at the optimum levels for maximum alfalfa productivity. The first cut was taken in January 2004 and the subsequent cuts were harvested at intervals of 30-35 days. Twenty cuts were obtained during the two growing years 2004 and 2005. Fresh weight and dry matter forage yield (FY and DY), plant height (H), number of tillers per square meter (T) and leaf to stem ratio (LSR) characteristics were studied. Data were tested for homogeneity before pooling. The combined analysis of variance was computed over two years. Stepwise multiple linear regression analysis was carried out as applied by Draper and Smith (1966). Phenotypic correlations were calculated according to Snedecor and Cochran

(1967). Path-coefficient analysis was calculated according to the procedures suggested by Dewey and Lu (1959).

## Results and discussion

The analysis of variance showed significant differences between years and among varieties for all traits studied. Varieties x years interaction was significant indicating that the varieties responded differently from year to year, and it is therefore essential to evaluate such varieties for a number of years and locations as mentioned by Bakheit (1988).

The combined analysis over the two years (Table 1) revealed that Syn<sub>2</sub> had significant higher fresh-weight yield (171.1 t ha<sup>-1</sup>) than the other genotypes. Moreover, Syn<sub>1</sub> (155.8 t ha<sup>-1</sup>) had no significant difference from Ismailia-94, which ranked third and was significantly different from Ismailia-1 and Siwa varieties. In terms of forage dry matter yield, the combined analysis indicates the same trend of significance: Syn<sub>2</sub> ranked first (46.9 t ha<sup>-1</sup>) and was significantly different from the other varieties, followed by Ismailia-94 (42.6 t ha<sup>-1</sup>) with no significant difference from Syn<sub>1</sub> (42.3 t ha<sup>-1</sup>), which ranked third; Ismailia-1 and Siwa had the lowest dry matter yields (38.7 and 37.1 t ha<sup>-1</sup>) and were significantly different from each other.

For measures of plant height, number of tillers and leaf to stem ratios, Syn<sup>2</sup> gave the highest values in each character, significantly different from the other entries in all traits.

The results support those of Abdel-Galil *et al.* (2000) who reported greater variability and dry matter yield among seven alfalfa cultivars at the New Valley than at Ismailia region.

Table 1. Combined 2-year results for fresh and dry matter yield (t ha<sup>-1</sup>), plant height (cm), number of tillers per square meter and leaf to stem ratio.

Var./Traits	Fresh yield (t ha <sup>-1</sup> )	Dry matter yield (t ha <sup>-1</sup> )	Height (cm)	Tillers	LSR (%)
Siwa	136.2	37.1	45.4	330.5	41.4
Ismailia-1	141.6	38.7	45.4	338.7	41.0
Ismailia-94	155.2	42.6	46.3	383.3	43.3
Syn <sub>1</sub>	155.8	42.3	45.6	365.2	42.2
Syn <sub>2</sub>	171.1	46.9	47.6	447.8	47.2
Mean	152.0	41.5	46.1	373.1	43.0
L.S.D.	2.354	0.590	0.429	12.59	1.11

Significant positive correlation were found between either fresh or dry yield and each of plant height and number of tillers (Table 2).

Table 2. Correlation matrix for all variables over two years.

Traits	Fresh yield	Dry matter yield	Plant height	Tillers
Dry matter yield	0.953**			
Plant height	0.762**	0.835**		
Tillers	0.653**	0.428**	0.318**	
L.S.R.	0.717**	0.730**	0.763**	0.351**

\*\*Significant at 0.01 level of probability.

These results are in line with the findings of Bakheit (1988) who reported positive correlation among plant height and either fresh and or dry forage yields in alfalfa. These results indicated the importance of selection for such traits to obtain high productive synthetic varieties or improved cultivars. The step-wise multiple linear regression analysis removed leaf to stem ratio variable (LSR) as it had low partial correlation and low F value.

Path-coefficient analysis was employed to determine the relative importance and direct and indirect effects for height and tillers as determiners of fresh forage yield. Results indicated that number of tillers revealed the most prominent direct effect on fresh forage yield (51.8%) followed by plant height (42.4%), while the indirect effects were 13.5% and 33.8% for these



traits respectively. Moreover, the tillers trait was found to be relatively important and influenced fresh forage yield by 25.9%, followed by plant height (21.3%) under the environmental conditions of this investigation (Table 3).

Table 3. Path-coefficient analysis and relative importance (R I %) of seasonal plant height (H) and number of tillers (T) upon fresh forage yield (FY) for five alfalfa varieties estimated over two years 2004 and 2005.

Case	Pathway of association			
	(H) vs.(FY)	(RI %)	(T) vs. (FY)	(RI %)
Correlation	0.7625		0.65288	
Direct effect	0.4241	21.3	0.51819	25.9
Indirect effect	0.3383	16.9	0.13469	6.7

## Conclusions

The alfalfa synthetics (Syn<sub>1</sub> and Syn<sub>2</sub>) investigated here showed suitability for the New Valley environment, giving a greater fresh weight herbage yield when compared with other varieties, despite having been developed under the Giza environment. Syn<sub>2</sub> population could be considered as a new promising population. The number of tillers and plant height characters influence the fresh forage yield trait. Therefore, plant breeders of alfalfa should consider these two characters in formulating their breeding programmes when selecting for highly productive varieties.

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# **Yield potential and stability performance of some Egyptian clover genotypes under different environments**

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## **Abstract**

A field study was conducted to evaluate the potential forage yield and phenotypic stability of 16 genotypes of Egyptian clover (*Trifolium alexandrinum* L.) at four regions of Egypt (Sakha, Gemmiza, Serw and Sids) during the 2003-04 and 2004-05 winter seasons. In terms of yield of herbage fresh weight, the genotypes Hatour, Sakha-4, Gemmiza-1, Narmer and Giza-6 exceeded the other genotypes, with no significant differences among each other, and were significantly different from the other genotypes. The genotypes Sakha-96, Gemmiza-1 and Hatour showed the highest phenotypic stability.

Keywords: Egyptian clover, phenotypic stability

## **Introduction**

Egyptian clover (*Trifolium alexandrinum* L.) is the main winter forage crop in Egypt. It is cultivated for animal feed and for its role in improving soil fertility. Development of varieties with high yielding ability and consistency requires that attention be given to the importance of stability performance for the genotypes under different environments, and their interactions. Eberhart and Russell (1966) suggested that the regression coefficients ( $b$ ) and deviations from regression ( $S^2d$ ) may be considered as two parameters for measuring the phenotypic stability. Abdel-Galil *et al.* (1998) reported that the Egyptian clover genotypes Sakha-4 and Serw-1 were desirable varieties for less favourable environments ( $b < 1$ ) but Giza-6, Giza15 and Helally were better adapted to favourable environments ( $b > 1$ ). This investigation aimed to determine the yield potential of 16 Egyptian clover genotypes and also to estimate their phenotypic stability performance under different environments.

## **Materials and methods**

A field experiment was carried out at Sakha, Gemmiza, Serw, and Sids Research Stations, representing different locations in the Delta and Middle Egypt, during the two successive seasons, 2003-04 and 2004-05, to evaluate the yield and stability performance of 16 Egyptian clover genotypes. Each season in each location was considered as an environment, so that the data obtained were considered as a response to eight environments. The Randomized Complete Block Design with four replicates was used, in which the plot size was  $6m^2$  and seeds at the rate of  $47.6 \text{ kg ha}^{-1}$  were hand drilled in rows 20 cm apart. The sowing dates were between 11 October and 18 October in both seasons at the four locations. The first cuts were obtained after 50 days from the sowing dates, and the subsequent cuts were taken at 25-30 days from the previous cut. Cultural practices were maintained at optimum levels to maximize productivity, and four cuts were obtained from each location in each season and fresh forage yields were recorded. Individual analysis of variance was applied for each location in each season and data were tested for homogeneity, then the combined analysis for the total cuts over seasons and locations was carried out according to Snedecor and Cochran (1989). Data were analysed using Mstat-C computer program (1986). The phenotypic stability parameters ( $b$  and  $S^2d$ ) were detected using the model described by Eberhart and Russell (1966).

## Results and discussion

Significant differences among the tested genotypes, years, locations and interactions were found, indicating that the tested genotypes were affected by the varying environments and there exists a wide variability among genotypes and environments. Significant interaction had presented some difficulty in identifying superior forage yielding clover genotypes over environments; thus it was found necessary to estimate the consistency of these entries for varying environments (Bakheit, 1985). The genotypes Hatour, Sakha-4 and Gemmiza-1 had the greatest yields of fresh weight (113.9, 113.0, and 112.3 t ha<sup>-1</sup> respectively) with no significant differences from Narmer, Giza-6, Sakha-96 and Hellaly (111.8, 110.9, 110.6, and 110.4 t ha<sup>-1</sup> respectively) and significant different from the other genotypes (Table 1).

In addition, the significance of the genotype x environment interaction indicated that the location had the major effects on the relative genotypic potential for fresh yield. This means that, for reliable evaluation of clover yield, it would be necessary to evaluate the genotypes with great emphasis on multi-location testing as reported by Abdel-Galil *et al.* (1998). Consequently, stability performance should be identified to get acquainted with the reaction and the response of each genotype to environmental changes. It could be stated from the values of regression coefficients (b) and deviation from regression mean squares (S<sup>2</sup>d) that the genotypes Sakha-96 (No.3), Gemmiza-1 (No.7) and Hatour (No.16) gave the most stable yields according to Eberhart and Russell (1966). However, the (b) values did not differ significantly from unity (b = 1) and (S<sup>2</sup>d) did not differ significantly from zero (S<sup>2</sup>d = 0) and they had higher fresh yield than the overall mean (Table 1).

Table 1. Overall means of fresh yield (t ha<sup>-1</sup>), regression coefficient (b) and deviation from regression mean squares (S<sup>2</sup>d) values for 16 genotypes of Egyptian clover (*Trifolium alexandrinum* L.).

No	Genotypes	Fresh yield (t ha <sup>-1</sup> )	b	S <sup>2</sup> d
1	<b>Sakha 3</b>	107.1	1.065	6.243*
2	Sakha 4	113.0	1.0004	2.691*
3	Sakha 96	110.6	0.9844	1.373
4	Helally	110.4	0.9887	12.84*
5	Giza 6	110.9	1.0225	6.384*
6	Giza 15	108.2	0.8959	1.319
7	Gemmiza 1	112.3	0.8769	1.358
8	Serw 1	108.1	0.9906	5.227*
9	Serw 2	108.0	0.964	4.131*
10	Sids Syn.	107.1	0.9907	1.469
11	Assiut Pop.	108.8	0.9985	1.889
12	Cairo 1	105.2	1.0648	3.449*
13	Cairo 2	109.0	1.0797	4.447
14	Cairo 3	104.0	1.0404	0.831
15	Narmer	111.8	1.0127	5.598*
16	Hatour	113.9	1.0248	1.539
Mean		109.4		
L.S.D. at 0.05%		2.930		

\*S<sup>2</sup>d values significantly varied from zero at 0.05 level of probability.

These genotypes could be considered as being stable, in terms of their fresh forage yield. The genotypes that gave a fresh-weight herbage yield in the range 104.0-109.0 t ha<sup>-1</sup> are also considered to be highly promising genotypes, as their potential yields are higher than the known typical values obtained by clover growers. Therefore, efforts should be directed to develop the stability performance for these genotypes under specific regions.

## Conclusions

Of the genotypes studied, Hatour, Sakha-4, Gemmiza-1, Narmer and Giza-6 gave the greatest herbage yields, and Sakha-96, Gemmiza-1 and Hatour are shown to be distinguished as stable genotypes where they gave the most stable yields, based on the stability parameters of Eberhart and Russell (1966).

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## Ability of grasses to compete in galega swards

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### Abstract

The suitability of various grasses to complement closed and productive mixtures with the long-term legume, fodder galega (*Galega orientalis* Lam.) was studied in field trials at the Research Station of Lithuanian University of Agriculture in 1999–2001. The trials were carried out on a sandy moraine humic horizon of a *Calcary–Epihypogleyic Luvisol*. The productivity, feed value and soil cover were found to be better for galega-grass swards than pure galega. It was found that a mixture with 50% *Festuca pratensis* and *Dactylis glomerata* had the highest ability to compete in swards with galega. Swards of galega mixed with *Poa pratensis* and *Lolium perenne* were the most productive. The largest yields were obtained at the first cuts each year. In all studied mixtures galega began to dominate grasses only in the third year. The most suitable grass components for the most productive galega mixtures were with 25% of *Phleum pratense* and 25% of *L. perenne*.

Keywords: fodder galega, grasses, swards, ecology, productivity

### Introduction

Biodiversity in agro-environmental fields could be improved by growing mixtures of different botanical composition. Mixed swards may improve soil cover in the sowing year and provide better, more stable and balanced chemical composition of yields over a longer period than pure-sown legumes or grasses. Chemical composition, especially sugar balance, is also better in legume–grass mixtures than in pure legume stands (Drikis, 1995). Legumes and perennial grasses maintain or increase natural fertility, agrochemical and microbiological properties of soil by accumulating biological nitrogen and organic matter (Szyszkowska *et al.*, 2004). It is generally recommended to grow legumes in mixtures with grasses (Halling *et al.*, 2002).

All these properties of legumes and their mixtures with grasses may give improved yields, and protein-rich, ecologically safe forage on ecologically sensitive soils. Legume-based agro-systems also provide environmental benefits in the medium term due to their ability to fix atmospheric N, thus contributing to decreased greenhouse gas emissions with a positive impact on climate change in the longer term (Hopkins and Wilkins, 2006).

The main aim of this research was to compare and select the most suitable long-term perennial grasses for developing stable, lasting, productive and chemically balanced swards with fodder galega (*Galega orientalis* Lam.).

### Materials and methods

In order to compare the productivity and suitability of the new fodder galega variety 'Vidmantai' in mixtures with different grasses, field trials were conducted at the Research Station of the Lithuanian University of Agriculture (LUA) in 1999–2001, on a sandy moraine loam humic horizon of a *Calcary–Epihypogleyic Luvisol* (LVg–p–w–cc). Binary mixtures of galega with long rooted meadow fescue (*Festuca pratensis* Huds.) cv. 'Dotnuvos I', timothy (*Phleum pratense* L.) cv. 'Gintaras II', cocksfoot (*Dactylis glomerata* L.) cv. 'Asta', thick sod-forming perennial ryegrass (*Lolium perenne* L.) cv. 'Veja' and smooth-stalked meadow grass (*Poa*

*pratensis* L.) cv. 'Danga' were studied. A randomized block design with four replications was used and the results are presented in Tables 1 and 2. The crops were sown without cover crops and cultivated without N fertilizer. The field was fertilized every autumn with P<sub>60</sub> and K<sub>60</sub>. A 3-cut system was applied manually from the beginning of the flowering stage. Competition was estimated as the underground mass share (%) of botanical groups in mixture (Freckleton and Watkinson, 2000). The chemical composition of dry matter (DM) was determined by near infra-red (NIR) computer analyser (PSCO/ISI IBM-PC 4250) according Wende forage analysis (Nauman and Bassler, 1983). The amount of metabolic energy (ME) and digestible protein (DP) were calculated according to the chemical composition of DM and coefficient of digestibility (CD). Botanical composition was determined from 0.25 m<sup>2</sup> of each replicate plot by weighing and calculating the share of each botanical group in the sward.

ANOVA was used for statistical analyses. The least significant difference (*LSD*<sub>05</sub>) method was used to evaluate differences between the crop yields, chemical composition and feed value at *P* < 0.05.

## Results and discussion

Grass-galega mixtures covered the soil evenly during the first year, in contrast to the pure galega crop. Meadow-grass and perennial ryegrass mixtures established rapidly and produced yields early in the sowing year as well as in the springtime (first cut) of the following years. The fast development of these mixtures could explain why swards were not invaded by unsown grasses or weeds (*Rumex crispus* etc.) for a long time. Tall grasses (meadow fescue and cocksfoot) showed the same persistence and competitiveness as the perennial ryegrass.

In agreement with Drikis (1995) the sward of pure galega did not reach the maximum potential of shoot development nor did it provide even cover of soil in the first and second years due to its slow initial development. Therefore, grasses (with the exception of timothy and meadow grass at 50% of seeding rate), dominated the botanical composition of swards during first two years (Table 1). The share of grasses comprised 46.3-58.3% of the botanical composition of these treatments. There was a tendency for the share and DM yield of galega to dominate at the second and third cut, except in the mixtures with cocksfoot.

Table 1. Botanical composition of swards in (species as %) of grass-galega mixtures.

Botanical composition, %	Pure galega	Galega/grass mixtures									
		Timothy		Meadow fescue		Cocksfoot		SS Meadow-grass		Perennial Ryegrass	
		50	25	50	25	50	25	50	25	50	25
Grass	0	50	75	50	75	50	75	50	75	50	75
Galega	100										
1st yr sward	95.8	19.4	18.6	54.2	24.5	55.5	34.7	52.5	30.9	53.1	22.4
Unsown grasses, %	1.7	2.3	1.9	2.0	1.8	1.9	1.8	2.6	2.0	2.1	2.2
Weed	2.5	3.2	2.3	2.9	2.4	2.8	2.9	3.7	2.4	2.3	2.3
2nd yr sward	94.2	46.3	18.4	53.6	26.1	56.3	33.9	49.9	25.1	51.6	22.7
Unsown grasses, %	2.1	2.8	1.9	2.5	2.2	2.7	2.3	3.8	3.7	2.0	1.8
Weed	3.7	3.9	2.8	3.8	3.4	3.6	3.6	4.2	3.9	2.6	2.5
3rd yr sward	93.6	42.3	16.1	48.4	24.0	49.2	30.8	41.8	20.2	46.7	18.4
Unsown grasses, %	3.7	3.0	2.4	2.6	2.4	3.0	2.7	4.1	3.9	2.6	2.3
Weed, %	2.7	3.3	3.0	4.0	3.6	3.9	4.0	4.7	4.5	3.1	3.0

The share of galega in the botanical composition increased and prevailed over the grasses in the third year due to complete shoot development. Galega started crowding out grasses and dominated in all treatments and its share in the third-year swards ranged from 50.8 to 84.9% (sown with 50% cocksfoot or 25% timothy, respectively) depending on the seed rate.

In the third year grasses comprised 41.8-49.2% (with 50% sown meadow-grass or cocksfoot, respectively) or 16.1-30.8% (with 25% sown timothy or cocksfoot). The content depended on the seeding rate and competitive ability of the grass. Typically, competition is a process



determined by life-strategy mechanisms and resource acquisition. Cocksfoot was the most aggressive grass with respect to galega. The following sequence represents the competitiveness of grasses in descending order: cocksfoot > meadow fescue > smooth-stalked meadow-grass > perennial ryegrass > timothy. The soil was completely covered in all treatments, so the amount of the unsown grasses and weeds constituted only 1.8-4.7%.

The content of DM, FU and DP per ha fluctuated depending on grass component and the respective yield in different treatments. The mixtures with perennial ryegrass (25%), timothy (25%) and pure galega were the most productive and produced 13.1; 12.8 and 12.7 t ha<sup>-1</sup> DM respectively. Galega mixtures with meadow-grass (50%) and meadow fescue (50%) produced significantly less yield than other mixtures, only 10.0 and 10.1 t ha<sup>-1</sup> DM.

Table 2. Productivity and feed value of fodder galega and its swards with grasses.

Indices	Galega	Timothy		Meadow Fescue		Cocksfoot		SS Meadow-grass		Perennial Ryegrass		
Grass, %	0	50	25	50	25	50	25	50	25	50	25	
Galega, %	100	50	75	50	75	50	75	50	75	50	75	LSD <sub>05</sub>
GM, t ha <sup>-1</sup>	61.7	57.1	62	49.1	54.4	53.2	57.6	50.8	54.2	51.2	63.2	6.6
DM, t ha <sup>-1</sup>	12.7	11.9	12.8	10.1	11	10.7	11.7	10	10.6	10.8	13.1	1.3
ME, MJ ha <sup>-1</sup>	11.2	10.8	11.3	9.5	9.9	9.8	10.7	9.5	9.7	9.9	11.8	6.79
DP, un. ha <sup>-1</sup>	2201	2120	2206	1955	2012	1997	2077	1961	2006	2076	2252	131

The greatest 3-year average yields of mixed swards were obtained from the first cut because of better agro-climatic conditions than at the time of the second and third cuts.

## Conclusions

Fodder galega is suitable for cultivating in mixtures with grasses. Mixed swards guarantee a more-even covering of soil in the sowing year and also provide higher, more stable and balanced chemical composition over a longer period than pure-sown galega or grasses.

The study found that the degree of aggressiveness of grasses with respect to galega was in the following decreasing order: smooth-stalked meadow-grass, meadow fescue, perennial ryegrass and timothy. Soil cover was complete in all treatments, so the amount of the unsown grasses and weeds constituted only about 2-4%.

The most suitable grass components to mix with fodder galega appeared to be timothy and perennial ryegrass.

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# Pasture establishment from different seed mixtures of grass and clover species under varied seeding conditions

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## Abstract

The yield of a new pasture depends on the relative proportions of plant species in the seed mixture, seeding rate and environmental conditions. We investigated how yield is affected by changing the proportions of two grasses (*Lolium perenne* L. and *Phleum pratense* L.) and two clovers (*Trifolium repens* L. and *Trifolium ambiguum* M. Bieb) in a seed mixture using a simplex design. Effects of changing the overall seeding rate (12 and 20 kg of viable seed ha<sup>-1</sup>) and level of N fertilizer (100 and 200 kg N ha<sup>-1</sup> y<sup>-1</sup>) were also examined. The design was repeated at two sites in Ireland. Response variables were annual herbage yields of sown and unsown (weeds) species in the first (2006) and second (2007) years after seeding. Results were consistent across sites and N levels. There was no difference between overall seeding rates in yields of sown species, but in 2006 there were more unsown species for the low than high seeding rate. In both years, total yields of mixtures were greater than monocultures, whereas yields of unsown species were lower in mixtures and grass monocultures than clover monocultures. Changing the relative proportions of species in the seed mixture affected species' yields, but *T. ambiguum* established poorly in all pastures.

Keywords: pasture establishment, seed mixtures, simplex design

## Introduction

Pasture seed mixtures sown in Ireland are mostly two-species mixtures of perennial ryegrass (*Lolium perenne* L.) and white clover (*Trifolium repens* L.). These species are suited to most of Ireland's environmental and management conditions. However, temporal and spatial instability of simple mixtures with only one grass and one clover species can contribute to variable livestock performance and supply of organic N. Other grass and clover species with different growth, persistence and nutritional characteristics, such as timothy (*Phleum pratense* L.) and Caucasian clover (*Trifolium ambiguum* M. Bieb), could be included in the seed mixture to improve the yield and quality of the sward. The yield and quality of multi-species pasture mixtures is likely to depend on the relative proportions of plant species included in the seed mixture, overall seeding rate and soil fertility. It is also likely to vary from one location to another. Furthermore, most temperate pasture species differ in their rates of seedling growth and development, which need to be considered when making decisions of time of sowing and composition of pasture seed mixtures (Black *et al.*, 2006). The objective of this study was to determine the effects of changes in the relative proportions of two grass and two clover species in a seed mixture, as well as overall seeding rate and N fertilization, on the yield and botanical composition of swards at two sites in Ireland.

## Materials and methods

Two grass species, *L. perenne*, cv. Spelga (Lper) and *P. pratense*, cv. Motim (Ppra), and two clover species, *T. repens*, cv. Avoca (Trep) and *T. ambiguum*, cv. Endura (Tamb) were used. Four monocultures of seed of each species and 11 seed mixtures with different proportions of each species were defined using a simplex design (Cornell, 2002). There were four mixtures dominated by each species (proportionately 0.7 of one species and 0.1 of each the other



species), six mixtures dominated by pairs of species (0.4 of each of two species and 0.1 of the other two species) and one mixture with each species equally represented (0.25 of each species). Treatments were repeated at two overall seeding rates (12 and 20 kg viable seed ha<sup>-1</sup>) at an N fertilizer level of 100 kg N ha<sup>-1</sup> y<sup>-1</sup>. The effect of a second level of N fertilizer (200 kg N ha<sup>-1</sup> y<sup>-1</sup>) was tested on the four monocultures, four mixtures dominated by each species, and the mixture with each species equally represented, at the two overall seeding rates. The 48 treatments were assigned randomly to plots (2 m × 5 m) at two sites in Ireland: Fermoy, County Cork (52°8'N, 8°16'W, 48 m above sea level) and Athenry, County Galway (AY; 53°17'N, 8°44'W, 40 m above sea level).

The plots were sown at Athenry on 27 July 2005 and at Fermoy on 15 August 2005. Pastures were managed by cutting with seven harvests in the first (2006) and second (2007) years after seeding. All plots at each site were harvested on the same day after 5-6 weeks of regrowth. Fertilizer N, P (35 kg ha<sup>-1</sup> y<sup>-1</sup>) and K (150 kg ha<sup>-1</sup> y<sup>-1</sup>) were applied after each harvest. Herbage dry matter (DM) yields of sown and unsown species were estimated at each harvest by cutting an area (Fermoy: 0.7 m × 5 m; Athenry: 1.28 m × 5 m) in each plot to a height of 4 cm using an Agria mower. The freshly cut samples were weighed and a sub-sample (200 g) dried at 40 °C (for 48 h) to determine DM yield. A second sub-sample was separated to determine the DM yields of each sown species and the unsown species fraction.

Data were analysed using a mixed effects model:  $Y_{ijkl} = \mu + M_i + R_j + N_k + S_l + \varepsilon_{ijkl}$ , where  $Y_{ijkl}$  is the yield of the *i*th mixture (*M*), *j*th seeding rate (*R*), *k*th N level in the *l*th site (*S*),  $\mu$  is the overall mean,  $M_i$ ,  $R_j$  and  $N_k$  are fixed effects,  $S_l$  is a random effect and  $\varepsilon_{ijkl}$  is the error.

## Results and discussion

In the first year after seeding, mean total yield was similar ( $P = 0.39$ ) between the two sites and averaged 8.85 Mg DM ha<sup>-1</sup>. There were no significant differences ( $P > 0.05$ ) between the two seeding rates in mean total yield and in yields of sown species, but there was a small decrease in mean yield of unsown species with increased seeding rate (-0.23 Mg DM ha<sup>-1</sup>,  $P < 0.05$ ). Mean total yield increased with N level (+0.55 Mg DM ha<sup>-1</sup>,  $P < 0.01$ ), which was largely accounted for by greater ( $P < 0.05$ ) yields of Lper and Ppra in monocultures rather than any differences in total yields of mixtures and clover monocultures (Figure 1a).

There were significant differences ( $P < 0.001$ ) between yields across seed mixtures (Figure 1a). For the monocultures, yields of Lper, Ppra and Trep were greater than Tamb ( $P < 0.001$ ), which established poorly in all pastures. Mean total yield of mixtures was greater than monocultures (9.52 vs. 7.51 Mg DM ha<sup>-1</sup>,  $P < 0.001$ ), which suggests a synergistic effect of mixing species. Mean yield of unsown species was lower ( $P < 0.001$ ) in mixtures (0.60 Mg DM ha<sup>-1</sup>) and grass monocultures (0.44 Mg DM ha<sup>-1</sup>) than clover monocultures (3.65 Mg DM ha<sup>-1</sup>), which suggest an antagonistic effect of mixtures on ingress of unsown species.

In the second year, mean total yield was greater ( $P < 0.001$ ) at Fermoy than Athenry (12.08 vs. 10.18 Mg DM ha<sup>-1</sup>). There were no significant differences ( $P > 0.05$ ) between the two seeding rates in mean total yield and in yields of sown and unsown species. Mean total yield increased with N level (+0.45 Mg DM ha<sup>-1</sup>,  $P < 0.01$ ) and was affected ( $P < 0.001$ ) by seed mixtures (Figure 1b). Monoculture yields were greatest for Ppra, intermediate for Lper and Trep and lowest for Tamb ( $P < 0.001$ ), which was almost absent from all established swards. The mean total yield of mixtures was greater ( $P < 0.001$ ) than monocultures (11.60 and 10.18 Mg DM ha<sup>-1</sup>) while there were fewer ( $P < 0.001$ ) unsown species in mixtures (mean of 0.62 Mg DM ha<sup>-1</sup>) and in the Ppra monoculture than in the Lper and clover monocultures.

Changing the relative proportions of species in the seed mixtures affected the yields of sown species in both years (Figure 1). There were positive linear associations ( $P < 0.001$ ) between proportions of species in the seed mixture and mean yields of each species, which were

strongest for Lper ( $R^2 = 0.79$ ) and weakest for Tamb ( $R^2 = 0.30$ ). However, in general, productive pastures were established and weeds were suppressed using all 11 seed mixtures.

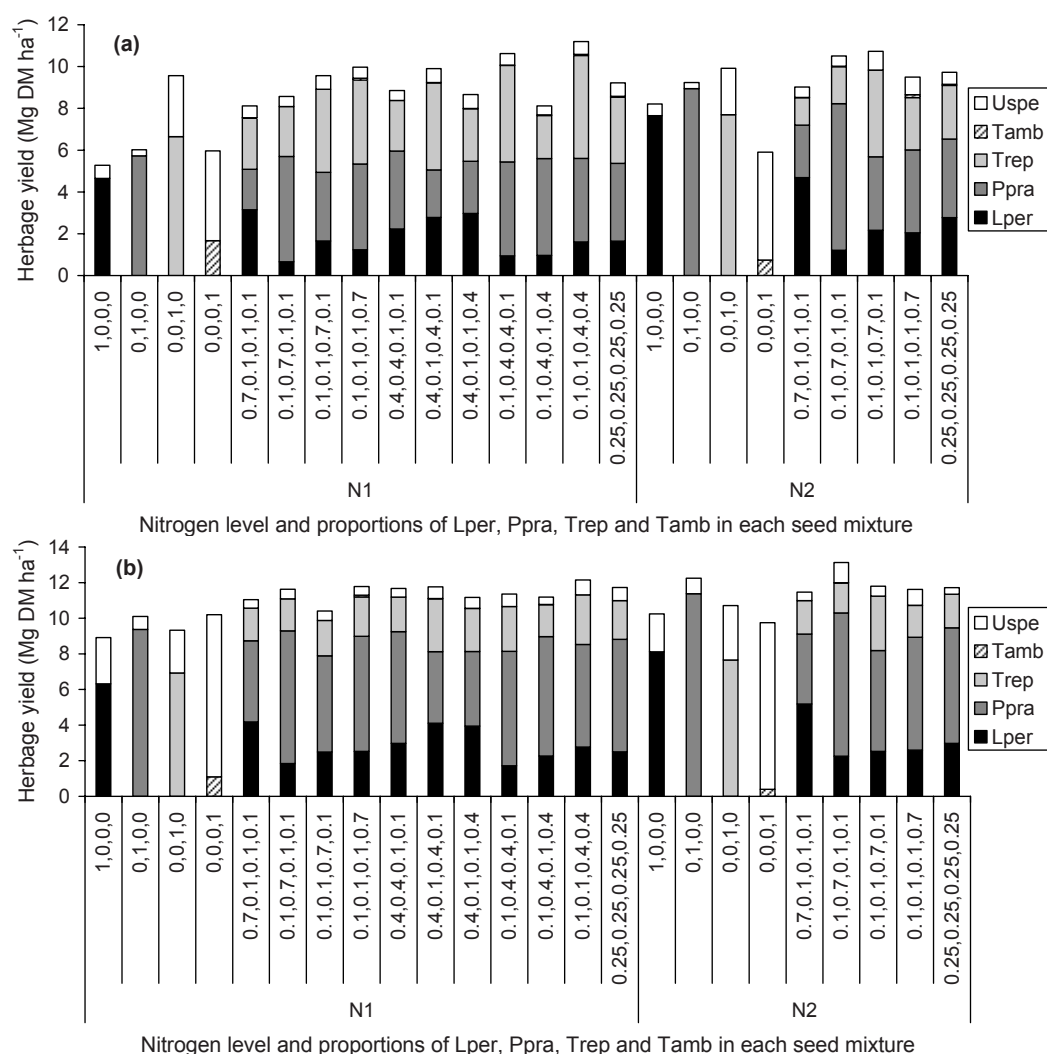


Figure 1. Mean accumulated annual yields of *Lolium perenne* (Lper), *Phleum pratense* (Ppra), *Trifolium repens* (Trep), *Trifolium ambiguum* (Tamb) and unsown (Uspe) species in the (a) first and (b) second year after sowing from seed mixtures with different species proportions and two levels of N fertilizer (N1 and N2). Data are means of two seeding rates and two sites.

## Conclusion

There was no difference between the two overall seeding rates in yields of sown species, but in 2006 the low seeding rate had more unsown species for the high rate. In both years, total yields of mixtures were greater than monocultures, whereas yields of unsown species were lower in mixtures and grass monocultures than clover monocultures. Changing the relative proportions of species in the seed mixture affected the yields of sown species, but *T. ambiguum* established poorly in all pastures. The results were consistent across both sites.

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# Leaf functional traits in *Brachypodium pinnatum* (L.) Beauv., *Bromus erectus* Hudson and *Dactylis glomerata* L. as indicators of resource availability in grassland communities

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## Abstract

*Brachypodium pinnatum*, *Bromus erectus* and *Dactylis glomerata* are very common species in European semi-natural grasslands but have different life strategies: *B. pinnatum* and *B. erectus*, unlike *D. glomerata*, are stress-tolerant competitors of xeric, resource-poor habitats. The study was made in spring and autumn of 2006, at two grassland areas (Regional Park of Monte Sole, Emilian Apennines) having similar climate conditions, but different resource availability. At each area Specific Leaf Area (SLA), Leaf Dry Matter Content (LDMC), Leaf Nitrogen Concentration (LNC) and Leaf Lignin Content (LLC) were measured in the three species. Variability of these functional traits was tested, by studying species, spatial and temporal variations, in order to investigate their value as indicators of different environmental conditions. The variations of LNC between seasons, and of SLA, LDMC and LLC between species, was highly significant. Different responses to environmental conditions in the species were confirmed by ranking of plant trait values. *D. glomerata* was characterized by the highest SLA and lowest LDMC and LLC values, fitting the expectations of fast-growing species. *B. pinnatum* and *B. erectus*, with lower SLA and higher LDMC and LLC values, tend towards the behaviour of slow-growing species. SLA and LDMC were found to be the major traits characterizing inter-site differences.

Keywords: Plant functional traits, *Brachypodium pinnatum*, *Bromus erectus*, *Dactylis glomerata*, grasslands

## Introduction

In semi-natural grasslands, nutrient resource availability is a determining factor influencing vegetation dynamics and species interactions of plant community structure. Species with different ecological strategies can coexist under given environmental conditions, but with different phenotypic adaptation in response to the variability of the resource availability. Three common species in the *Mesobromion*-type grasslands of the North Apennines were chosen for our study: *Dactylis glomerata*, which dominates the mesic nutrient-rich end of the gradient, and *Brachypodium pinnatum* and *Bromus erectus*, which tend to be the most competitive at the mesic, nutrient-poor end of the gradient (Corcket *et al.*, 2003). A functional trait approach, describing different trade-offs related to plant strategies and competitive ability of dominant species, has been shown to explain species occurrence along the environmental gradient. Positive associations are found between Specific Leaf Area (SLA, leaf area to dry mass ratio), Leaf Nitrogen Concentration (LNC), the rate of photosynthetic capacity and rapid biomass production (Garnier *et al.*, 2001). Low SLA species typically have high values for Leaf Dry Matter Content (LDMC, leaf dry mass to saturated fresh mass ratio) and Leaf Lignin Content (LLC), which lead to efficient conservation of nutrients (Aerts and Chapin, 2000). The aim of our study was to test the variability of SLA, LDMC, LNC and LLC across time, site and species, in order to investigate whether the species ranking for these leaf trait values could be indicators to characterize plant responses to environmental conditions.

## Materials and methods

The study was conducted in two protected areas, Zannini and Stanzano, in the Regional Park of Monte Sole (6,476 ha), in the south-west Emilian Apennines (Italy). The areas have similar climate (in 2006, annual precipitation was 706 mm, nearly 22% of which fell in September, and mean annual temperature was 11.5°C). Both sites are at 400 m a.s.l., but have different soil texture (silt loam at Zannini, 44% sand, 54% silt and 2% clay, and silt at Stanzano, 12% sand, 85% silt and 2% clay), aspect (respectively south and south-west) and slope (respectively 25° and 16°). At each of three sites in the Zannini area and two sites in the Stanzano area, three plots were selected for leaf sampling. Collections were carried out in spring (second half of May) and in autumn (first half of October). For *B. pinnatum*, *B. erectus* and *D. glomerata*, SLA and LDMC were measured on 10 leaves from different vegetative individuals randomly chosen in each plot, healthy and fully developed, following the protocol of Garnier *et al.* (2001). Foliar surfaces were measured in the laboratory by a Li-COR Area Meter model 3100 (Lincoln, NB, USA). LNC and LLC were measured on further leaves by selecting the youngest, fully expanded leaves for each species, weighing 10 g, collected from the same plots. Samples were oven dried at 60 °C and ground for nitrogen content analysis by the Kjeldahl method and for fibrous fractions (NDF, ADF, ADL) by the method of Goering and Van Soest (1970). Spatio-temporal differences in SLA, LDMC, LNC and LLC were evaluated using one-way ANOVA.

## Results and discussion

Figure 1 shows results of box plots for testing trait variations between seasons, species and sites. Differences between the two sampling seasons in 2006 were not significant for SLA, LDMC and LLC, but were for LNC ( $P < 0.001$ ), with values increasing from spring ( $1.4 \pm 0.1 \text{ g kg}^{-1} \text{ dm}$ ) to autumn ( $2.1 \pm 0.2 \text{ g kg}^{-1} \text{ dm}$ ) (Fig. 1a, d, g and l). Consistently lower LNC values in spring than in autumn may be the consequence of N-deficiency produced by low water availability during the spring drought, but species rankings for other traits do not seem to have been influenced. Interspecies variations were significant for SLA ( $P < 0.001$ ), LDMC ( $P < 0.001$ ) and LLC ( $P = 0.018$ ), but not for LNC (Fig. 1b, e, h and m). SLA values for *D. glomerata* ( $22.1 \pm 2.6 \text{ m}^2 \text{ kg}^{-1}$ ) were on average higher than for *B. pinnatum* ( $19.2 \pm 2.2 \text{ m}^2 \text{ kg}^{-1}$ ) and *B. erectus* ( $16.74 \pm 2.7 \text{ m}^2 \text{ kg}^{-1}$ ), unlike LDMC values which were lower in *D. glomerata* ( $230.2 \pm 24.3 \text{ mg g}^{-1}$ ) than *B. erectus* ( $273.6 \pm 24.6 \text{ mg g}^{-1}$ ) and *B. pinnatum* ( $316.0 \pm 40.8 \text{ mg g}^{-1}$ ). Average LLC value of *D. glomerata* ( $5.7 \pm 1.8 \text{ g kg}^{-1} \text{ DM}$ ) was lower than *B. erectus* ( $6.2 \pm 1.2 \text{ g kg}^{-1} \text{ DM}$ ) and *B. pinnatum* ( $7.6 \pm 1.2 \text{ g kg}^{-1} \text{ DM}$ ). A fast rate of resource acquisition is expected to be associated with higher SLA but lower LDMC and LLC, characterizing competitive ability of fast-growing species, as in *D. glomerata*. Intermediate SLA values in *B. pinnatum*, and lower SLA values in *B. erectus*, confirm a generally higher competitive ability for *B. pinnatum* than *B. erectus*, but a higher tolerance to poor conditions for *B. erectus*. Moreover, leaves with higher LDMC and lignin content, as in *B. pinnatum*, tend to be relatively tough and long-lived, more resistant to physical damage and more efficient in the retention of nutrients, than leaves with low LDMC and LLC, as in *D. glomerata*. Differences between sites were significant only for SLA and LDMC ( $P = 0.018$  and  $P = 0.032$ ) with average SLA values higher in leaves from Zannini ( $20.4 \pm 3.2 \text{ m}^2 \text{ kg}^{-1}$ ) than Stanzano ( $17.6 \pm 2.8 \text{ m}^2 \text{ kg}^{-1}$ ) and the inverse trend for LDMC (respectively  $258.8 \pm 44.3 \text{ mg g}^{-1}$  and  $295.6 \pm 42.7 \text{ mg g}^{-1}$ ) (Fig. 1c, f, i, n). The major constraint on plant growth at Stanzano is likely to be the high percentage of silt (85%) in the soil. Small pore diameter poses a serious limitation to water conductivity and soil aeration, and causes impedance to root penetration and nutrient acquisition. Here, species tend to have lower SLA and higher LDMC to counterbalance leaf growth by an increased necessity for conserving resources.

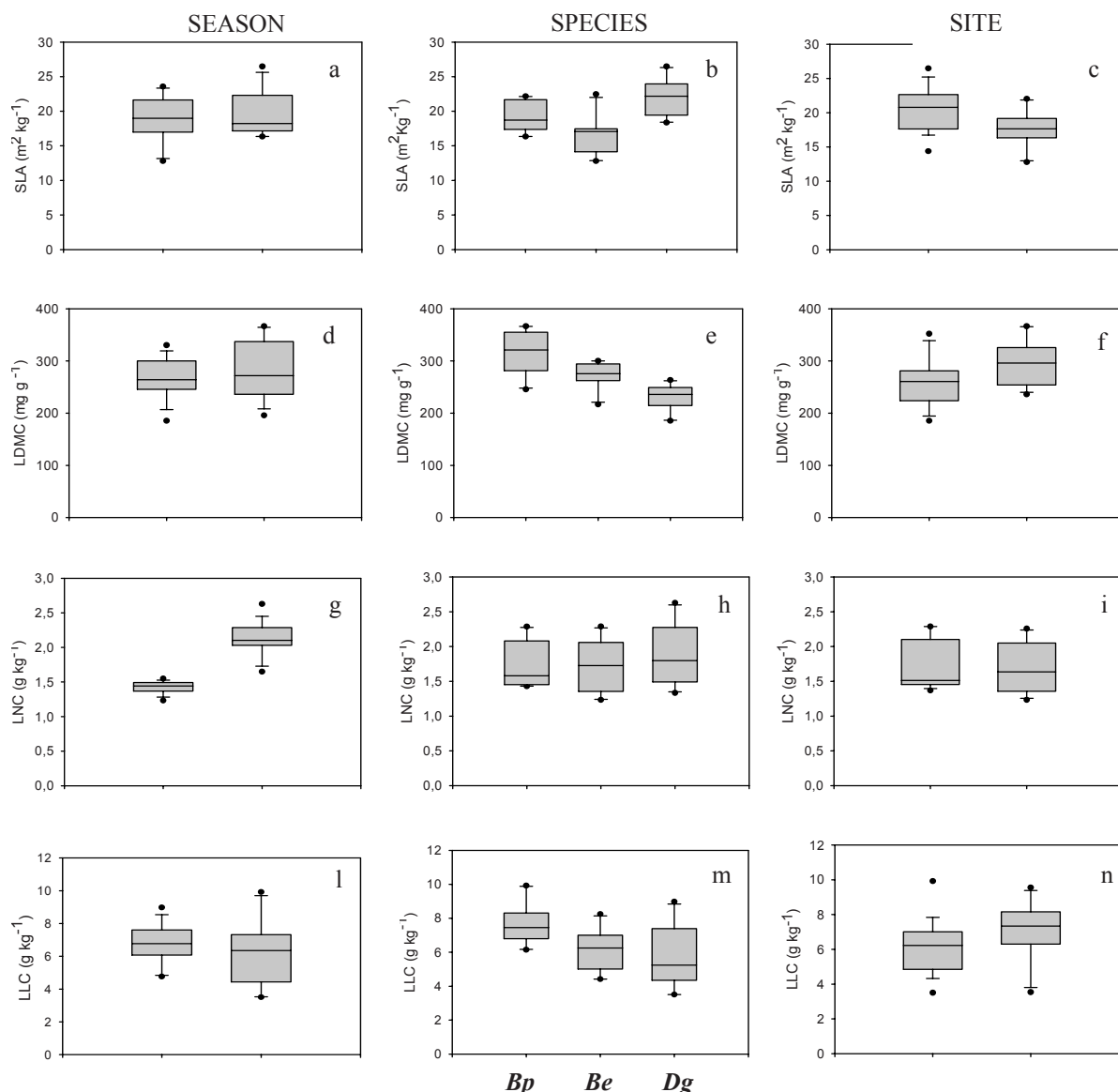


Figure 1. Box-plots of spatio-temporal variations for SLA (Fig. 1a,b,c), LDMC (Fig. 1d,e,f), LNC (Fig. 1g,h,i), LLC (Fig. 1l,m,n) in *B. pinnatum* (*Bp*), *B. erectus* (*Be*), *D. glomerata* (*Dg*).

## Conclusions

Our study proposes SLA and LDMC are the major traits to characterize different environmental conditions between sites, taking into account that rankings in plant functional trait values may be modulated by the specific ecological strategy of each species. Since there is large between-season variation in values of LNC, and LLC shows variation between species but not between sites, we suggest that these traits have little value for our purposes.

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# Effects of cutting management on the floristic composition of two permanent Italian ryegrass (*Lolium multiflorum*) meadows

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## Abstract

With the aim to evaluate the effects of cutting frequency on botanical composition, two trials were carried out on intensively cultivated permanent ryegrass meadows in the northern Veneto plain. The meadows were managed with 6, 5 or 4 cuts per year for three consecutive years, and before each cut a botanical survey was carried out. The results from the third year, which highlight that cutting frequency had a remarkable effect on the botanical composition, are reported. The increase in cutting frequency led to a significant decrease in the proportion of Italian ryegrass.

Keywords: *Lolium multiflorum*, Italian ryegrass, permanent meadows, cutting frequency

## Introduction

Nowadays farmers need to improve their forage quality in order to reduce the quantity of fodder products brought on to the farm. A way to tackle this issue is to shorten the period between cuts; consequently, the forage is produced by younger plants which normally are poor in structural carbohydrates. In addition, the presence of smaller plants, like clovers, that have a high content of crude protein, could be increased. Two plot trials were conducted on two different intensively cultivated Italian ryegrass (*Lolium multiflorum*) meadows in the upper Veneto plain. The aim was to study the effects of the management on meadow characteristics. In each trial three different harvest times were compared for three years. In the work described here, results from the assessments of botanical composition during the third year of experimentation are presented.

## Materials and methods

In February 2005 two different permanent meadows were selected in the upper Veneto plain in Italy as typical examples of lowland grasslands in this area (Rodaro *et al.*, 1997; 2000). The two meadows differ in terms of soil type: the first (Site 1), located in Carmignano di Brenta (45°38'N, 11°42'E; 46 m a.s.l.) is on a silt clay loam soil with good permeability; the second (Site 2), located in Gazzo Padovano (45°17'N, 11°53'E; 36 m a.s.l.) is on clay loam soil (less permeable and thus more prone to retention of surface water). At both sites the experimental design was a randomized block with four replicates. In each trial there were 9 treatments, obtained from the factorial combination of 3 cutting frequencies and 3 levels of fertilization (organic and mineral), that were compared. Plot size was 4.00 m x 6.00 m = 24.00 m<sup>2</sup>, the central part of which (3.00 m x 6.00 m = 18.00 m<sup>2</sup>) was used as experimental area. The three different cutting frequencies were as follows: 50 days, corresponding to 4 cuts year<sup>-1</sup>; 38 days, corresponding to 5 cuts year<sup>-1</sup>; and 30 days, corresponding to 6 cuts year<sup>-1</sup>. These frequencies were chosen in order to start and complete the annual cutting season simultaneously, in each case. During the period 2005-2007, the first cut was made on all plots on a date that coincided with *Lolium multiflorum* sprouting (about 10th of May). In accordance with accepted practice of the study area, flood irrigation was performed 5 times per year.

Immediately before each cut, the botanical composition was assessed according to the Braun-Blanquet (1964) method. Subsequently, in order to have a feasible comparison among results, values were averaged in the three following groups: 1) data of first and second cut for all cutting frequencies, 2) data of the third and fourth cut belonging to the 38-days and 30-days cutting frequencies, and 3) data of the fifth and sixth cut belonging to the 30-days cutting frequency. Finally, ANOVA was performed considering the per cent abundance of each plant species. Below, for the sake of brevity, only the botanical composition assessments for 2007 are presented; in addition, the reported results and discussions are limited to the effects of different cutting frequencies, since, in each case, the cutting frequency x fertilization interactions were not significant.

## Results and discussions

During the third year a significant decrease of *L. multiflorum* was observed as a consequence of increased frequency of cutting (Table 1).

Table 1. Species abundance (percentage of ground cover) at the time of each cut in the third year, referred as average between two cuts or for single cut.

Plots: cuts per year	4	5	6	sig	4	5	6	sig	4	5	6	sig
Reference cuts	1-2	1-2	1-2		3	3-4	3-4		4	5	5-6	
<b>Site 1: Carmignano di Brenta</b>												
<i>Lolium multiflorum</i>	46.3	30.8	16.7	**	23.5	15.4	8.5	*	4.6	2.7	1.3	**
<i>Poa trivialis</i>	5.9	3.6	4.4		-	-	-		-	-	-	
<i>Dactylis glomerata</i>	-	-	0.0		-	-	0.0		-	-	0.1	
<i>Setaria glauca</i>	-	2.1	2.7		13.8	22.1	26.7	**	8.7	11.5	13.2	
<i>Digitaria sanguinalis</i>	-	-	-		0.6	4.1	1.5	*	4.1	12.2	18.4	**
<i>Echinochloa crus-galli</i>	0.3	-	-		8.3	1.6	1.0	*	9.1	0.2	1.4	*
<i>Sorghum halepense</i>	0.1	-	-		0.3	-	-		0.1	-	-	
<i>Trifolium repens</i>	7.6	12.4	15.8	*	10.0	14.9	21.4	**	8.8	16.1	18.8	*
<i>Trifolium pratense</i>	0.2	0.4	0.6		0.6	0.3	0.3		0.4	0.1	0.2	
<i>Stellaria media</i>	9.0	13.0	16.8	*	1.4	0.2	0.1		28.6	24.9	11.0	*
<i>Veronica persica</i>	1.0	1.8	1.4		-	0.1	0.1		4.1	8.2	4.7	
<i>Capsella bursa-pastoris</i>	1.2	3.0	3.9	*	-	-	-		-	0.1	0.0	
<i>Crepis vesicaria</i>	0.0	0.4	0.5		-	-	-		-	-	-	
<i>Erigeron annuus</i>	0.5	1.2	1.7	*	-	0.5	0.5		0.0	-	0.1	
<i>Sonchus oleraceus</i>	-	0.6	0.1		-	-	0.0		-	0.7	0.1	
<i>Rumex obtusifolius</i>	5.0	7.9	12.3	*	5.7	8.2	12.9	*	5.8	7.5	11.1	**
<i>Taraxacum officinale</i>	4.9	6.9	13.6	*	5.1	11.1	15.6	*	5.5	8.2	15.9	*
<i>Rorippa silvestris</i>	0.0	0.3	1.4		0.4	0.2	1.3		0.3	0.3	0.7	*
<i>Convolvulus arvensis</i>	14.6	11.0	3.2	**	24.4	16.7	6.6	**	12.9	5.7	2.1	*
<i>Ranunculus acris</i>	1.5	0.7	1.1		3.3	1.1	0.4	**	3.3	1.3	0.4	*
<i>Ranunculus repens</i>	0.1	-	-		0.4	-	0.0		0.3	0.1	-	
<i>Plantago lanceolata</i>	0.0	-	-		-	-	-		0.1	0.1	0.0	
<b>Site 2: Gazzo Padovano</b>												
<i>Lolium multiflorum</i>	54.3	49.2	36.9	**	32.9	24.4	17.9	**	7.1	4.7	5.5	
<i>Alopecurus utriculatus</i>	1.2	3.1	4.9	*	-	-	-		-	-	-	
<i>Poa trivialis</i>	14.5	15.8	24.5	*	-	-	-		-	-	-	
<i>Setaria glauca</i>	0.6	1.0	-		20.3	32.1	36.0	**	38.6	43.4	49.7	*
<i>Digitaria sanguinalis</i>	-	-	-		-	1.6	-		1.3	3.8	8.5	**
<i>Echinochloa crus-galli</i>	-	-	-		1.2	1.0	0.3		2.3	0.9	0.8	*
<i>Trifolium repens</i>	7.0	7.3	9.9		14.4	9.4	17.3		15.9	11.5	13.9	
<i>Veronica persica</i>	-	-	0.0		-	0.1	0.0		-	0.5	0.1	
<i>Taraxacum officinale</i>	4.4	10.4	17.8	*	5.0	10.6	19.5	*	5.0	10.7	10.0	*
<i>Ranunculus repens</i>	16.8	13.4	6.0	*	25.7	20.1	8.9	*	29.5	22.8	11.2	*
<i>Ranunculus acris</i>	0.3	-	-		0.4	0.4	0.0		0.4	0.4	-	
<i>Potentilla reptans</i>	0.1	0.2	-		-	0.5	0.1		-	0.8	0.4	

\*:  $P < 0.05$ ; \*\*:  $P < 0.01$

At Site 1 in the first two cuts, per cent abundance of *L. multiflorum* decreased from 46.3% to 30.8% passing from 4 cuts year<sup>-1</sup> to 5 cuts year<sup>-1</sup>, and declined to 16.7% where 6 cuts year<sup>-1</sup> were taken. At the same time, at Site 2, per cent abundance decreased from 54% to 49% and to 37%. Similarly, in the following cuts, despite the decline in the absolute cover values for *L. multiflorum* due to the specific biological cycle of the plant, the effects of the different number of cuts per year on the per cent abundance of this grass persisted with a similar trend. Compared with *L. multiflorum*, other plant species showed a contrasting response; among these, four botanical families were of particular interest: 1) *Poaceae*, with *Poa trivialis* and *Alopecurus utriculatus*, which were present only in spring, *Setaria glauca* which was more abundant in summer and autumn, like *Digitaria sanguinalis*; 2) *Fabaceae*, with *Trifolium repens*, which at Site 1 showed this trend in all cuts; 3) Other perennial species such as *Taraxacum officinale*, *Rumex obtusifolius* and *Rorippa silvestris*; 4) annual or biennial species belonging to other botanical families, like *Capsella bursa-pastoris*, *Erigeron annuus*, *Crepis vesicaria* and finally *Stellaria media* which was present only in spring (first and second cuts). A trend similar to *L. multiflorum* was observed for *Echinocloa crus-galli* and *Sorghum halepense* (with reference to *Poaceae* family), and *Convolvulus arvensis*, *Ranunculus repens* and *Ranunculus acris* for perennial species, while in the autumn season only *Stellaria media* of the rest of the botanical families.

If we refer to the findings of a previous work (Rodaro *et al.*, 2000) which evaluated the contribution to the forage value by the species which had the greatest effect, we can assume that a high frequency of cutting could lead to a worsening of the meadow quality as a result of the decline of *L. multiflorum* abundance; causing, at the same time, an increase of species such as *R. obtusifolius*, *T. officinale* and, especially, *S. glauca*. A partial improvement of grassland quality can be expected as a consequence of the increased abundance of *T. repens*, but this effect is probably not enough to compensate for the effects of a decrease in *L. multiflorum*.

## Conclusions

Different cutting frequencies in intensively cultivated permanent ryegrass meadows led to a significant change in botanical composition. In general, increasing the frequency from 4 to 6 cuts per year caused a decrease of the abundance of *L. multiflorum* and other perennial species. At the same time, an increase in abundance of other species characterized by a poor forage quality, such as *Setaria glauca*, *Digitaria sanguinalis*, *Taraxacum officinale* and *Rumex obtusifolius*, occurred.

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# Study on long-term meadow productivity and botanical composition in response to different liming and fertilization

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## Abstract

A study was conducted in 2003-2006 to determine the role of liming and fertilization on long-term meadow productivity and botanical composition. Under  $P_{60}K_{60}$  fertilization the dry matter (DM) production of the permanent meadow was greatest ( $2.32\text{--}4.43\text{ t ha}^{-1}$ ) when the soil  $pH_{KCl}$  was adjusted to 5.6-6.0. When  $N_{120}P_{60}K_{60}$  fertilization was applied, the greatest DM yield ( $3.0\text{--}6.33\text{ t ha}^{-1}$ ) was obtained when the soil  $pH_{KCl}$  was  $\geq 6.1$ . Rather different meteorological conditions occurred during the growing seasons of the 4-year trial period, especially in the amount of precipitation, which resulted in large DM yield differences within the same pH level and changes in botanical composition. In the drier and warmer year of 2006 the proportion of grasses decreased while that of forbs increased. The most favourable year for legume growth was 2004, when precipitation during vegetation period was not high (279 mm), but distributed more uniformly. Liming to different pH levels had no considerable influence on sward botanical composition. However, fertilization with  $N_{120}P_{60}K_{60}$  led to an increase in the proportion of grasses, while the amount of forbs decreased by half and legumes almost died out (0.90-3.46%).

Keywords: liming, fertilization, DM yield, botanical composition

## Introduction

Soil pH is one of the factors influencing meadow productivity and persistence. Permanent meadow grasses yield the best on soils where pH is adjusted to  $\geq 6.0$  pH and maintained at that level (Daugėlienė, 2002). Czech researchers (Kralovec and Lipavsky, 2000) have suggested a higher value (pH to 6.5) to which soils should be limed in order to guarantee good quality of the fodder. However, liming affects pasture yield formation less than mineral fertilizers, especially nitrogen (Daugėlienė, 2002; Palmborg *et al.*, 2004; Butkuvienė and Butkutė, 2005; Gałka *et al.*, 2005). Nitrogen (N) fertilization effect is high but it is profitable only up to the limit of response when 1 kg N gives 10 kg herbage dry matter (DM)  $\text{ha}^{-1}$  of extra yield (Daugėlienė, 2002). N fertilization affects changes in botanical composition (Daugėlienė, 2002; Alibegovic-Gribs *et al.*, 2005). For better legume growth there is a requirement for available P and K, and fertilization with  $P_{80}K_{80}$  increases legume amount in the swards (Gutauskas *et al.*, 2003). White clover proportion also increases when soil pH becomes more alkaline. However, legume variation is higher than for grasses and forbs; it is cyclical and depends on a complex of factors (Gutauskas, 2003). In this study sward DM yield and botanical composition under the influence of modified soil pH and mineral fertilization were investigated over four years.

## Materials and methods

The research was carried out during 2003-2006 on a meadow which had had 12-15 years of use, 10 years as a pasture and the last years as a hayfield meadow. The soil was a *Haplic Luvisol (LVh)*. A trial with four different soil pH treatments was imposed on two background levels of fertilization, in a randomized design with three replicates of each treatment, and a plot size of 18 x 2.8 m. The four different soil  $pH_{KCl}$  levels (5.1-5.5; 5.6-6.0; 6.1-6.5 and 6.6-

7.0) and two background fertilization rates ( $P_{60}K_{60}$  and  $N_{120}P_{60}K_{60}$ ) were formed by liming and fertilization. The following fertilizers were used: ammonium nitrate (34.7% N), bone-dust (30.7%  $P_2O_5$ ) and potassium-magnesium (24-30%  $K_2O$ ). The PK fertilizers were applied in early spring before the start of grass growth, and N fertilizer was applied in two equal doses after the first and second cuts. Swards of different botanical composition were formed: a white clover (*Trifolium repens* L.) and meadow grass (*Poa pratensis* L.) sward in  $P_{60}K_{60}$ , and a uniform meadow grass sward in the  $N_{120}P_{60}K_{60}$ . Three cuts were taken by a self-propelled haymaker Hege 211 when grasses were at the ear-emergence stage, and sward dry matter (DM) yield and its botanical composition were determined. Precipitation during the vegetation period in each year of the trial was as follows: 337 mm in 2003, 279 mm in 2004, 553 mm in 2005 and 203 in 2006 mm.

## Results and discussion

Despite the changing standpoint to agriculture and its influence on contamination level the productivity of agricultural plants remains the key outcome of agricultural activity.

As indicated in Table 1, agronomic measures such as liming and fertilization influenced the meadow productivity. Under  $P_{60}K_{60}$  fertilization the permanent meadow was the most productive (2.32-4.43 t DM ha<sup>-1</sup>) when soil pH<sub>KCl</sub> was adjusted to 5.6-6.0. When  $N_{120}P_{60}K_{60}$  fertilization was applied, the highest meadow yield (3.0-6.33 t DM ha<sup>-1</sup>) was obtained when soil pH<sub>KCl</sub> was  $\geq 6.1$ . Liming had a greater effect on swards treatments fertilized with  $P_{60}K_{60}$  than with  $N_{120}P_{60}K_{60}$ .

During the trial period, on the meadow treatments fertilized with  $P_{60}K_{60}$  the DM yield increased by 34-43% at pH 5.6-6.0, by 12-36% at pH 6.1-6.5, and by 10-39% at pH 6.6-7.0, as compared with the yield at pH 5.1-5.5. However, fertilization with  $N_{120}P_{60}K_{60}$  resulted in only a 3-6% increase in DM yield, and even in a 1-9% decrease when the productivity of the meadow on soils with different pH levels was compared with the pH 5.1-5.5 level. The decrease in DM yield was obtained in 2005 and 2006, years when the precipitation during the vegetation period was the highest (553 mm) and the lowest (203 mm) respectively.

Table 1. Effect of liming and fertilization on long-term meadow productivity.

pH <sub>KCl</sub>	DM yield, t ha <sup>-1</sup>							
	$P_{60}K_{60}$				$N_{120}P_{60}K_{60}$			
	2003	2004	2005	2006	2003	2004	2005	2006
5.1-5.5	3.10	2.72	3.06	1.67	5.94	5.38	4.91	3.13
5.6-6.0	4.43	3.73	4.09	2.32	5.91	5.38	4.61	2.83
6.1-6.5	3.48	3.69	3.78	2.27	6.19	5.61	5.07	3.23
6.6-7.0	3.72	3.77	3.56	1.85	6.33	5.72	4.85	3.00
LSD <sub>05</sub>	1.03	1.79	0.92	0.64	0.80	0.46	0.45	0.32

Rather different meteorological conditions during the 4-year trial period, especially the amount of precipitation, resulted in quite great DM yield differences within the same pH level and changes in botanical composition (Table 2). In the drier and warmer year of 2006 the amount of grasses decreased and forbs increased. The most favourable year for legume growth was 2004 (with 26.60-33.20% legumes at  $P_{60}K_{60}$  and 1.80-3.40% at the  $N_{120}P_{60}K_{60}$  treatment) when the amount of precipitation during the vegetation period was not high (279 mm), but distributed more uniformly. The reduction in the proportion of forbs was also greater in 2004 than in the other years of the trial.

Average data for 2003-2006 showed (Table 2), that on meadow, fertilized with  $P_{60}K_{60}$ , liming increased the proportion of grasses by 2.78-7.70% and legumes by 0.53-6.47%, and considerably decreased the proportion of forbs by 7.42-10.42%. Liming to different pH levels had no sufficient influence on sward botanical composition under  $N_{120}P_{60}K_{60}$  fertilization.

However, fertilization with  $N_{120}P_{60}K_{60}$  led to an increase in the proportion of grasses, while the amount of forbs decreased by half and legumes almost died out, compared to  $P_{60}K_{60}$  fertilized meadow. The results showed that the proportion of white clover increased when soil pH became more alkaline. Nevertheless, the variation in the legume proportion was the highest, when compared with grasses and forbs under both fertilization backgrounds. Compared with the  $P_{60}K_{60}$  fertilized meadow, the variation in the proportion of grasses under  $N_{120}P_{60}K_{60}$  fertilization was lower, while that of legumes varied by almost the same coefficient of variation and forbs varied by a greater amount.

Table 2. The variation of sward botanical composition under different liming and fertilization.

Soil pH <sub>KCl</sub>	Grasses		Legumes		Forbs	
	average amount %	variation coefficient %	average amount %	variation coefficient %	average amount %	variation coefficient %
$P_{60}K_{60}$						
5.1-5.5	41.01 ± 3.82	18.65	16.67 ± 3.49	41.85	42.33 ± 3.30	15.61
5.6-6.0	47.90 ± 3.52	14.72	17.20 ± 4.90	57.00	34.91 ± 5.29	30.30
6.1-6.5	43.79 ± 3.60	16.46	23.14 ± 4.59	39.65	33.07 ± 3.21	19.41
6.6-7.0	48.71 ± 3.98	16.33	19.39 ± 4.35	44.92	31.91 ± 3.98	24.93
$N_{120}P_{60}K_{60}$						
5.1-5.5	78.85 ± 4.02	10.21	1.96 ± 0.39	40.27	19.19 ± 4.32	45.03
5.6-6.0	76.97 ± 4.02	10.44	1.51 ± 0.31	41.13	21.52 ± 3.86	35.87
6.1-6.5	79.69 ± 3.95	9.91	2.41 ± 0.32	26.21	17.90 ± 4.14	46.29
6.6-7.0	78.04 ± 5.48	14.04	2.77 ± 0.40	28.93	19.94 ± 4.94	49.59

## Conclusions

The findings showed that meadows fertilized with  $P_{60}K_{60}$  with ~ 20% of legumes in the sward were the most productive when soil was limed to pH<sub>KCl</sub> of 5.6-6.0. Fertilization with  $N_{120}P_{60}K_{60}$  resulted in an increase in the proportion of grasses, the amount of forbs decreased by half and legumes almost died out. When fertilizing with  $N_{120}P_{60}K_{60}$ , soils should be limed to ≥ 6.1 pH in order to reach the highest productivity of the meadows.

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# Nitrogen fixation in clover grasslands of varied plant species richness

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## Abstract

Plant species diversity influences the intensity of inter-specific competition for soil resources. To investigate how plant species richness and composition influence N<sub>2</sub> fixation, the perennial clovers *Trifolium hybridum* L., *T. pratense* L., and *T. repens* L. were grown in experimental grassland plots at one site in Sweden and one in Germany. The species richness varied from monocultures up to 12 or 16 species per plot. The amount of N<sub>2</sub> fixed in each *Trifolium* species was measured according to the <sup>15</sup>N natural abundance and <sup>15</sup>N isotope dilution methods using three non-N<sub>2</sub>-fixing reference species. N<sub>2</sub> fixation ranged from close to 0 up to about 25 g N m<sup>-2</sup> year<sup>-1</sup> based on shoot biomass above 5 cm. The highest values were measured in species-poor communities where the legume(s) contributed most to the total biomass in the plots. Due to a consistently high reliance on N<sub>2</sub> fixation, the amount of N<sub>2</sub> fixed per area was strongly correlated with biomass of the three *Trifolium* species. Species richness had no consistent effect on N<sub>2</sub> fixation.

Keywords: <sup>15</sup>N methods, biodiversity, legumes, N<sub>2</sub> fixation, species richness, *Trifolium*

## Introduction

N<sub>2</sub> fixation in perennial forage legumes is influenced by several factors, including companion species, N fertilization, cutting regime, and weather conditions. Competition for both light and nutrients is intensified in species-rich plant communities (e.g. Scherer-Lorenzen *et al.*, 2003; Jumpponen *et al.*, 2005), but little is known about how N<sub>2</sub> fixation is influenced by the species richness of neighbouring vegetation. The aim of the work presented here was therefore to examine the effects of plant species richness on N<sub>2</sub> fixation in the three perennial *Trifolium* species: *T. hybridum* L. (alsike clover), *T. pratense* L. (red clover) and *T. repens* L. (white clover). Our hypothesis was that increasing species richness would have a positive influence on N<sub>2</sub> fixation per plant, expressed as the amount of N<sub>2</sub> fixed per sown seed.

## Materials and methods

The *Trifolium* species were grown in monocultures or in mixtures of varied plant species composition (up to 12 or 16 perennial plant species, both grasses and herbs) in experimental grassland plots located in Umeå, Sweden (63°49'N, 20°17'E) and Bayreuth, Germany (49°55'N, 11°35'E), established in spring 1996 as parts of the European BIODPTH project (Mulder *et al.*, 2002; Scherer-Lorenzen *et al.*, 2003). Because of reoccurring freeze-thaw damage causing plant death during winters at the Swedish site, the Swedish experimental plots were repaired by re-sowing in spring 1999 and in spring 2000. There was no fertilizer application in the period 1996-2001.

Each year, in mid-August at the Swedish site and in late June and mid-September at the German site, plant biomass samples above 5 cm were dried and weighed for dry matter (DM) determination of individual species. The four growing seasons 1996, 1998, 2000 and 2002 at the Swedish site, and the first harvest in 1997 at the German site were studied. Samples of *Trifolium* spp. and three non-N<sub>2</sub>-fixing reference species, both grasses and non-leguminous herbs, were ground in a ball mill and analysed for <sup>15</sup>N abundance and N concentration. In samples from the period 1996 to 2000, the proportion of legume N derived from N<sub>2</sub> fixation, pNd<sub>fa</sub>, was calculated according to the NA method (Amarger *et al.*, 1979). In early June 2002, a majority of the plots at the Swedish site were fertilized with <sup>15</sup>NH<sub>4</sub><sup>15</sup>NO<sub>3</sub> (5 atom % <sup>15</sup>N excess) corresponding to 5 g N m<sup>-2</sup>. In samples from 2002, pNd<sub>fa</sub> was calculated according to the isotope dilution (ID) method (Fried and Middleboe, 1977). Amounts of N<sub>2</sub> fixed per m<sup>2</sup> and year (Nfix) were calculated as:

$$\text{Nfix} = \text{harvested DM (g DM m}^{-2} \text{ year}^{-1}) \times \text{N concentration (g N g DM}^{-1}) \times \text{pNd}_{\text{fa}}$$

To account for the decreasing number of seeds sown per m<sup>2</sup> of each individual species with increasing species richness, we also expressed Nfix as the amount of N<sub>2</sub> fixed per seed by dividing Nfix for each *Trifolium* species with the number of sown seeds of that species in each plot. More detailed information about the experimental procedures will be presented in Carlsson *et al.* (Manuscript).

## Results and discussion

At the German site and in unfertilized plots at the Swedish site, the studied *Trifolium* species relied mainly on N<sub>2</sub> fixation across all levels of species richness (pNd<sub>fa</sub> was on average 0.7). In contrast, when the plots at the Swedish site were N fertilized, pNd<sub>fa</sub> was lower in all three *Trifolium* species (on average 0.5), and pNd<sub>fa</sub> often increased with increasing species richness (not shown). Such unusually low pNd<sub>fa</sub> values (Carlsson and Huss-Danell, 2003) might be due to the late and single harvest date in combination with the N fertilization.

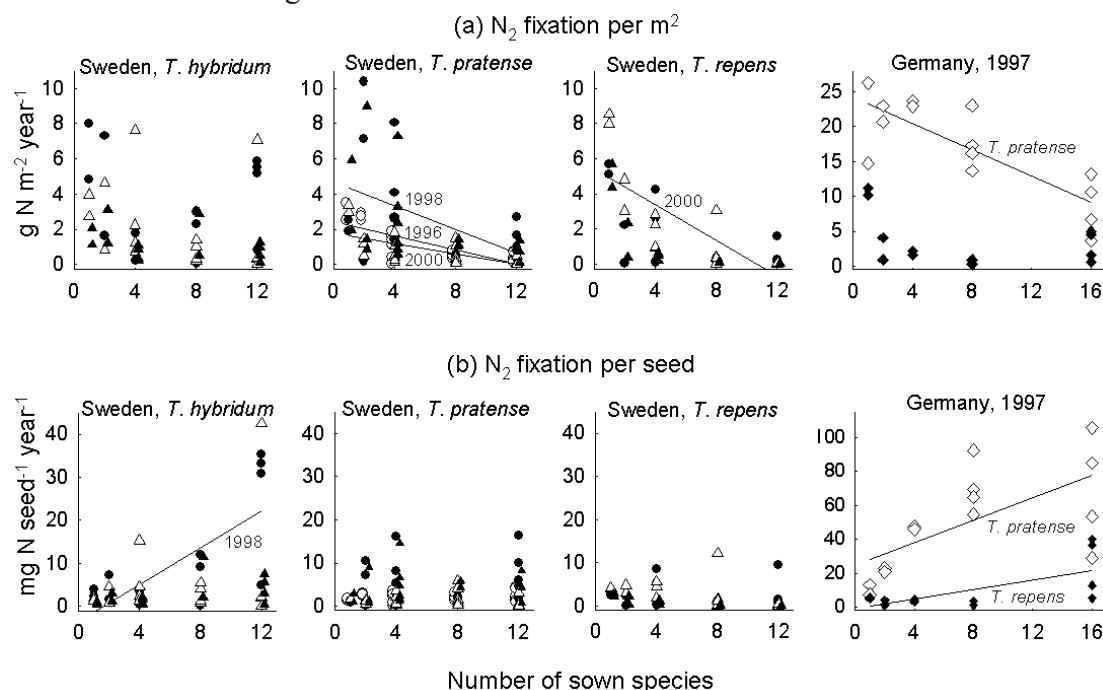


Figure 1. N<sub>2</sub> fixation in relation to the sown number of species. Open circles, Sweden 1996; filled circles, Sweden 1998; open triangles, Sweden 2000; filled triangles, Sweden 2002 (fertilized with 5 g N m<sup>-2</sup>); open diamonds, Germany *T. pratense*; filled diamonds, Germany *T. repens*. Each point represents one experimental plot. Regression lines indicate significant linear regression ( $P < 0.05$ ). Note different scales on y-axes for the Sweden and Germany.



The amounts of N<sub>2</sub> fixed per m<sup>2</sup> and year at the German site were often more than twice the amounts at the Swedish site, especially in *T. pratense* (Figure 1a). In *T. pratense* and *T. repens*, N<sub>2</sub> fixation per m<sup>2</sup> and year decreased with increasing species richness (Figure 1a). In addition, N<sub>2</sub> fixation per m<sup>2</sup> in each *Trifolium* species was positively correlated with its own contribution to total community biomass, but negatively correlated with biomass proportion of tall grasses (not shown). This negative correlation was likely due to light competition by tall grasses in diverse communities (Jumpponen *et al.*, 2005), which potentially limited legume biomass production and thereby the amount of N<sub>2</sub> fixed.

The negative species richness effect on amounts of N<sub>2</sub> fixed per area could be an effect of the decreasing proportion of individual species with increasing species richness that was inherent in the experimental design. Thus, we cannot with certainty distinguish whether Nfix was influenced mainly by species richness effects or by competition from a few competitive species. However, when the decreasing proportion of individual species with increasing species richness was accounted for by calculating Nfix per seed (Figure 1b), there was a positive species-richness effect at the German site for both *T. pratense* and *T. repens*, which is in line with our hypothesis. In Sweden, on the other hand, this effect was found only in *T. hybridum* in 1998 (Figure 1b). A positive species-richness effect on Nfix per seed might be caused by reduced intra-specific competition in species-rich communities. The reasons for different responses in *T. pratense* and *T. repens* at the German and Swedish sites are not clear, but factors such as differences in harvest time and identities of neighbouring species may have influenced the outcome.

## Conclusions

This study is the first to quantify effects of plant species richness on N<sub>2</sub> fixation in legumes. Clover biomass was the most important factor controlling the amount of N<sub>2</sub> fixed per area. If corrected for legume abundance, the amount of N<sub>2</sub> fixed per sown seed was either not affected or positively influenced by species richness. There was thus no consistent species richness effect on N<sub>2</sub> fixation.

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## Site effects on species productivity and dynamics: results from COST 852 in Wales

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### Abstract

An experiment quantifying diversity-function relationships in agro-ecosystems (COST 852) was established at a range of sites in Europe, including two sites in Wales. These were located in Aberystwyth (AB: lowland), and in Bronydd Mawr (BM: upland). The species and varieties within species were identical in the two sites, as were all major management factors. The species used were two grasses (*Lolium perenne* L. (perennial ryegrass) and *Dactylis glomerata* L. (cocksfoot)) and two legumes (*Trifolium pratense* L. (red clover) and *T. repens* L. (white clover)). In addition to the core COST 852 experiment, both sites also included an extra treatment whereby the genetic heterogeneity of the legumes was greatly increased by constructing mechanical mixtures comprising seed of many cultivars/populations. The results highlighted substantial differences in plant responses between the upland and lowland sites. The two contrasting environments produced two sets of quite different communities at the end of the experiment, and these were arrived at through very different patterns of species dynamics. However, in both sites, mixtures significantly outperformed monocultures in terms of yield of sown species over three years, exemplifying the agronomic benefits of species diversity. There was no consistent effect of legume genetic heterogeneity on community structure or function.

Keywords: legumes, grasses, productivity, species dynamics

### Introduction

Relationships between mixture diversity and function have been under-researched in an agronomic context, but a recent study carried out at 28 European sites (as part of COST Action 852) attempted to rectify this using methods that allowed assessment of the effects of species identity, density, richness and evenness in multi-species mixtures (Kirwan *et al.* 2007). Two of the 28 sites were located in Wales: Aberystwyth (AB), a lowland location (altitude 30 m.a.s.l.), and Bronydd Mawr (BM), an upland site (altitude 330 m.a.s.l.). This paper compared the development and composition of agronomic mixtures at the two sites, using models to test the persistence and effects of sown diversity. In addition, the effect of increased genetic heterogeneity of the legumes was measured.

### Materials and Methods

Communities of two legume and two grass species were constructed using a simplex design to create fifteen experimental communities with systematically varying evenness (relative abundance): monocultures, and mixtures with one dominant, two co-dominant species, or of maximum evenness ('centroid', equal proportions of four species). The experiment was sown in August 2002 in AB and in August 2003 in BM. The species and cultivars within species were identical in the two sites, as were sowing rates, cutting height (5 cm) and fertilizer applications (90 kg N ha<sup>-1</sup> total in three equal applications). The species used were two

grasses (G1: perennial ryegrass cv. Fennema, and G2: cocksfoot cv. Cambria) and two legumes (L1: red clover cv. Merviot, and L2: white clover cv. Alice). In addition to the core COST 852 experiment, both sites also included nine additional communities (four monocultures; four with one dominant species; a centroid) in which the genetic heterogeneity of both legume species was greatly increased by constructing seed mixtures made up of 11-14 commercial cultivars, plus some unselected 'gene pool' germplasm (Collins *et al.*, 2004). This is termed the 'broad genetic base' treatment (B), and the single cultivar treatment is termed 'narrow genetic base' (N). All experimental communities were replicated at a low and high level of overall initial biomass (low being 60% of high), leading to a total of 48 communities. Three years' data from each site were analysed. This analysis considered both measures of ecosystem function (annual yield of sown species and weeds) and changes in community structure (species relative proportions). The modelling approaches used to address these questions are described in Kirwan *et al.* (2007) for function, and Connolly and Wayne (2005) for structure.

**Functional response:** The modelling approach used allowed us to assess: (i) differences among species identity effects (individual species performances) and the impact of the genetic base treatment on this; (ii) whether there was an effect of increased species diversity; (iii) the impact of the genetic base treatment on the species diversity effect; (iv) whether the diversity effect was strong enough to produce transgressive over-yielding (TO) i.e. whether the mixture outperformed the best performing monoculture; and (v) whether all of these effects changed through time and across sites.

**Structural response:** Community dynamics were analysed using a relative growth rate difference (RGRD) model (Connolly and Wayne, 2005). If all species gained at the same per-unit rate, then community composition remained unchanged over time, whilst if the RGR of one species was greater than others then its relative proportion increased. This modelling approach tested whether the changes in community composition differed between the B and N treatments. The composition was predicted from the models for communities that began as centroids (equal species proportions) and this was compared between sites and across genetic base treatments.

## Results and discussion

**Functional response:** Results for Year 3 predicted from the model are shown in Figure 1.

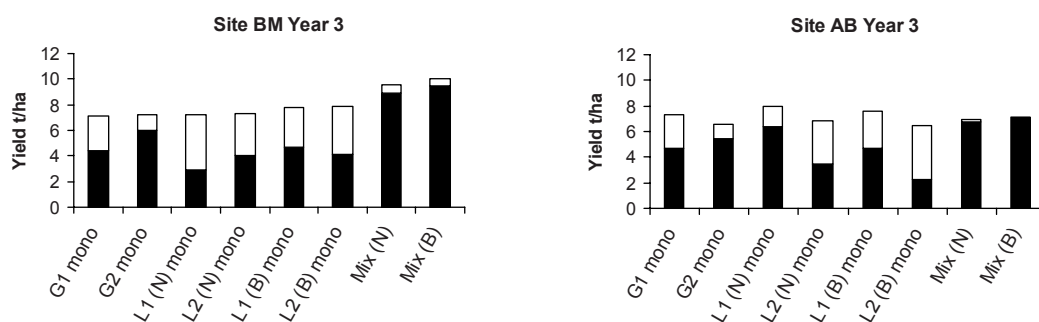


Figure 1. Predicted yields ( $\text{t ha}^{-1}$ ) of sown species (shaded) and weeds (unshaded) in monoculture and centroid communities in Year 3 derived from model [1] across sites. N and B are the narrow and broad genetic base treatments respectively.

One striking feature of the results was the difference between sites in the weed content of the communities, with plots in BM generally containing more weeds. This difference diminished by Year 3, as monocultures in site AB became invaded by weeds. In both sites and in all three years the yield of unsown species was much lower in the centroid mixtures than in



monocultures, exemplifying the positive effect of species diversity on resistance to weed invasion. The fact that this effect persisted for the three years of the experiment in both sites provides clear evidence for the long-term benefits of increased sward diversity. The response of red clover differed markedly between sites, with substantially higher yields of this species in AB than in BM in all communities. Yields of the other three species in the experiment were not as site-dependent.

In the N treatment in site AB TO occurred for annual yield in Year 2 and for the three-year cumulative yield. In the B treatment TO occurred for annual yield in Year 3 and for the three-year cumulative yield in 4 out of 5 mixtures. Thus, in the AB site there was a tendency for TO to become apparent in the later stages of the experiment. In contrast, TO occurred earlier and more frequently in both treatments in BM. In the N treatment at this site there were significant occurrences of TO in every year of the experiment as well as in the three-year cumulative yield. In the B treatment, TO occurred in 5 out of 5 mixtures in BM in all three years. The magnitude and generality of the TO response observed at the BM site strongly suggests that even modest increases in agronomic species diversity can contribute significantly to forage production in grassland systems in the uplands.

Overall differences in yield between the genetic base treatments were not significant except in AB in Year 3, where the red clover N treatment monoculture outyielded the B treatment ( $P < 0.054$ ), and in BM in Year 3 where the reverse was the case ( $P < 0.049$ ).

Structural response: There were significant differences between species' RGRs in most species pairs, particularly in Years 1 and 2. The genetic base treatment had a diminishing effect on RGRD as the experiment progressed, whereas site effects were significant for most species pairs up to the end of Year 3. There was a large difference between the sites in the 'end-point' composition of the centroids, with the AB site mixture containing a very large proportion of cocksfoot in both the N and B treatments by the end of Year 3. In contrast, both N and B centroid mixtures in BM comprised more equal proportions of the four species.

In this experiment we purposely avoided introducing extra variation between the sites by sowing the experiment with the same cultivars and by imposing the same management criteria. Despite this, the two contrasting environments produced two sets of quite different communities at the end of the experiment, and these were arrived at through very different patterns of species dynamics. These site effects could be caused by variation in biotic (e.g. pest and disease load; soil microbial population identity and abundance) and abiotic factors (climate; soil nutrient status and quality) or interactions between these. Some simple measures of the site environments are available: AB is consistently about 2 °C warmer than BM; the latter has more days with frost during winter and spring and has markedly more rainfall. Measurements of soil fertility at the start of the experiment showed that concentrations of P and K were lower in BM than in AB (in BM the soil P concentration was below optimum), and soil pH was also slightly lower in BM. The relative importance of these factors is not known as yet. Overall, the results highlight the importance of variety x environment interactions in determining the species composition of pastures for agricultural use. The results highlight the importance of species diversity in pastures, both for persistent productivity and weed suppression, particularly in sites that experience challenging environmental conditions.

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# Seasonal dynamics of dandelions (*Taraxacum officinale* L.) in permanent grassland

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## Abstract

The objectives of our research were to study the variation of dandelion (*Taraxacum officinale* L.) in a long-term pasture. Dandelions had very favourable background conditions to spread since they also dominated beyond the borders of the experiment. The experimental data, covering 11 years, show that the amount of dandelions in the pasture had been almost consistently decreasing from the first year up to the ninth year of use. During the growing season the amount of dandelions in the sward varied significantly. The highest content of dandelions was found in the dry matter yield of the first grazing, while the smallest content in that of the second grazing. Liming and fertilization decreased the amount of dandelions. The occurrence of dandelions was positively affected by soil pH, with fewer dandelions in the soil with a pH<sub>KCl</sub> of 6.6-7.0 than with a pH<sub>KCl</sub> of 5.1-5.5. Soil pH and fertilization caused no significant effect on the content of potassium in dandelions. The lowest concentration of potassium was estimated in the second grazing at the background fertilization of N<sub>0</sub>P<sub>60</sub>K<sub>60</sub>.

Keywords: liming, fertilization, dandelions, potassium, grazings

## Introduction

Climate change has a great long-term effect for the botanical composition of grassland. Dandelion (*Taraxacum officinale* L.) is a common grassland forb and has a high potential to invade newly established swards. Dandelion could increase the dry matter yield in grass/clover swards (Isselstein, 2000). The dominating forb species in Czech grasslands is also dandelion (Pavlu *et al.*, 2000). In extensively used German grasslands that are not fertilized with nitrogen, dandelions occupy about 25% of the grassland area (Baade *et al.*, 2000). The application of mineral fertilizers has seriously degraded the botanical diversity of grassland communities (Chapman, 2001). If the plots were grazed with animals, the content of dandelions would probably be reduced, and the grass content would be higher (Lunnan, 2001). We tested analysis of seasonal dynamics of dandelions in a permanent pasture during 11 years with four levels of liming and two levels of fertilization.

## Materials and methods

The soil of the experimental site is a sod podzolic Hapli-Endohypogleyic Luvisol (IDg4-p) light loam over medium loam, and has a top-soil pH<sub>KCl</sub> of 5.2, available P<sub>2</sub>O<sub>5</sub> of 108 mg kg<sup>-1</sup> and K<sub>2</sub>O of 142 mg kg<sup>-1</sup>. Lime was applied before pasture sowing. Limestone rate was calculated according to the titration curves neutralizing the soil with 0.033 N CaCl<sub>2</sub> solution. A grass-clover mixture, consisting of 35% *Trifolium repens* L., 40% *Phleum pratense* L. and 25% *Poa pratensis* L. was sown. The sward was fertilized annually in spring with 60 kg ha<sup>-1</sup> of both P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O. Nitrogen (N<sub>120</sub>) fertilizer was split-applied in two times after the first and second grazing. The treatments were replicated 4 times and grazed 4 times by a herd of dairy cows. The amount of dandelions in the samples was measured after separation as dry matter weight. DM yield was determined on the basis of total DM amount per plot and

calculated as DM yield ha<sup>-1</sup>. The data of DM yield were statistically analysed by the STAT-ENG according to (Tarakanovas, 1999).

## Results and discussion

The seasonality of dandelion occurrence depended on grazing time, soil pH<sub>KCl</sub> and nitrogen fertilization (Table 1). On the background of N<sub>0</sub>P<sub>60</sub>K<sub>60</sub>, the greatest content of dandelions in the DM yield of herbage in the first grazing, when dandelion was at mass flowering stage, was determined. The lowest content of dandelion was identified in the yield of the second grazing, since during the period from the first grazing to the second grazing the seed of dandelion did not emerge because dandelion seed does not have strictly expressed dormancy period. In the yield of the third grazing at soil pH<sub>KCl</sub> levels of 5.1-5.5, the content of dandelion was almost the same as in the first grazing, and at soil pH<sub>KCl</sub> levels from 5.6 to 7.0, the content of dandelion was significantly lower. An increase in the content of dandelion in the yield of the third grazing can be explained by seed emergence due to the improved light regime, since the height of herbage of this grazing, as well as the yield, were much lower than those of the first grazing. Moreover, some of the dandelion plants that had not flowered in spring started flowering, and some dandelion plants started flowering for the second time. In the herbage of the fourth grazing the content of dandelion declined again; however, its content was the same as in the second grazing. On the background of N<sub>120</sub>P<sub>60</sub>K<sub>60</sub>, at all soil pH<sub>KCl</sub> levels the content of dandelion was lower than that on the background of N<sub>0</sub>P<sub>60</sub>K<sub>60</sub>. The variation of dandelion content during the growing season was the same as on the background of N<sub>0</sub>P<sub>60</sub>K<sub>60</sub>; however, the distribution of dandelion amount was different. The significant difference is the fact that the declined dandelion content in the herbage of the second grazing did not restore, in any case, to the level of the first grazing. The grassland quality is influenced by the species composition and the changes occurring during the growing season (Fisher *et al.*, 1993). Nitrogen, promoting growth of grasses, improved botanical composition of the swards, since dense grass sward suppressed the spread of dandelion. Variation of dandelion amount in the pasture sward was very high; however, with decreasing soil pH<sub>KCl</sub>, the variation declined.

Table 1. Dynamics of the amount of dandelions in separate grazings as influenced by soil pH and fertilization.

Grazings:	Amount of dandelions % averaged over 11 years				Average amount of dandelions %	
	1	2	3	4	Minimum	Maximum
Soil pH <sub>KCl</sub>	N0P60K60				Minimum	Maximum
5.1-5.5	29.2 ± 3.83	19.1 ± 3.32	26.3 ± 3.89	20.2 ± 4.17	12	46
5.6-6.0	30.0 ± 4.27	18.7 ± 2.60	25.0 ± 3.75	19.0 ± 4.21	11	54
6.1-6.5	33.2 ± 3.93	18.5 ± 2.84	25.5 ± 3.64	19.5 ± 3.51	16	62
6.6-7.0	27.9 ± 2.74	15.5 ± 2.03	22.2 ± 2.54	17.4 ± 3.19	14	50
Soil pH <sub>KCl</sub>	N120P60K60				Minimum	Maximum
5.1-5.5	24.4 ± 2.42	11.2 ± 1.35	14.9 ± 1.69	13.2 ± 2.31	11	38
5.6-6.0	25.0 ± 3.31	11.1 ± 1.35	15.8 ± 2.19	12.1 ± 2.01	10	39
6.1-6.5	24.0 ± 1.81	12.6 ± 1.37	14.6 ± 1.41	11.7 ± 1.78	11	38
6.6-7.0	24.4 ± 2.01	9.9 ± 1.02	14.7 ± 0.93	10.0 ± 1.49	11	32

Table 2. The concentrations of potassium in separate grazings of dandelions as influenced by soil pH and fertilization.

Grazings:	Concentration of potassium % averaged over 11 years				Average concentration of potassium %	
	1	2	3	4		
Soil pH <sub>KCl</sub>	N0P60K60				Minimum	Maximum
5.1-5.5	2.74 ± 0.09	2.33 ± 0.15	2.85 ± 0.19	2.95 ± 0.16	1.52	3.79
5.6-6.0	2.79 ± 0.14	2.56 ± 0.16	2.86 ± 0.20	2.51 ± 0.16	1.52	3.59
6.1-6.5	2.73 ± 0.13	2.33 ± 0.16	2.80 ± 0.19	2.94 ± 0.18	1.46	3.84
6.6-7.0	2.70 ± 0.12	2.38 ± 0.15	2.82 ± 0.17	2.91 ± 0.17	1.41	3.65
Soil pH <sub>KCl</sub>	N120P60K60				Minimum	Maximum
5.1-5.5	2.55 ± 0.14	2.48 ± 0.17	2.38 ± 0.13	2.61 ± 0.14	1.60	3.48
5.6-6.0	2.50 ± 0.16	2.56 ± 0.17	2.52 ± 0.10	2.51 ± 0.16	1.52	3.52
6.1-6.5	2.49 ± 0.18	2.63 ± 0.17	2.54 ± 0.09	2.67 ± 0.13	1.61	3.55
6.6-7.0	2.45 ± 0.17	2.56 ± 0.16	2.54 ± 0.09	2.64 ± 0.13	1.51	3.42

The average content of potassium in pasture grass did not reach the limit of 3%. However, the highest content of potassium in all soil pH<sub>KCl</sub> and fertilization levels exceeded the last-mentioned limit (Table 2). The concentration of potassium did not exceed the permissible limit (3%) in those pastures where dandelions make up to 25% of sward botanical composition (Daugėlienė and Butkuvienė, 2005). The lowest concentration of potassium was found under the N<sub>0</sub>P<sub>60</sub>K<sub>60</sub> background in dry matter yield at the second grazing, because the leaves of dandelions were prevailing. There were no significant differences between potassium contents at the particular grazing of N<sub>120</sub>P<sub>60</sub>K<sub>60</sub> background, when the nitrogen promoted the growing of grasses and limited the development of dandelions.

## Conclusions

The spread of dandelion in a permanent pasture during the growing season was reduced by systematic grazing, liming (pH<sub>KCl</sub> 6.6-7.0) and fertilization (N<sub>120</sub>P<sub>60</sub>K<sub>60</sub>).

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# Potential of fodder legumes under intensive farming conditions in Flanders

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## Abstract

Farmers are encouraged by the Flemish Government to cultivate clover (*Trifolium* species) and lucerne (*Medicago sativa* L.) and grass/clover to produce more farm grown proteins and to decrease the use of mineral fertilizers. Monocultures of red clover (*Trifolium pratense* L.), white clover (*Trifolium repens* L.), perennial ryegrass (*Lolium perenne* L.), Italian ryegrass (*Lolium multiflorum* Lam.) and grass/legume mixtures were compared at 3 levels of N-fertilization: 0, 105 and 265 kg N ha<sup>-1</sup> in order to investigate the effect on botanical composition, yield, forage quality and nitrate residue in the soil. In the monocultures of grasses, weeds significantly decreased with fertilization while weeds tended to increase with fertilization in the legume monocultures. The use of 105 or 265 kg N ha<sup>-1</sup> y<sup>-1</sup> on grass/legume mixtures increased yield by 1.0 and 1.3 Mg ha<sup>-1</sup> respectively. No relationship was found between the energy content and the level of N fertilization. The protein content was more affected by type of monoculture or mixture than by applied N-level. The levels of nitrate residue in the soil were very acceptable for all the treatments; they were highest for the legume monocultures and lowest for the grasses.

Keywords: clover, lucerne, ryegrass/legume, yield, quality, nitrate residue

## Introduction

In Flanders farming moves towards less intensive agriculture mainly due to the environmental policy for restricting nitrate leaching. In this situation of a reduced N fertilization, legumes could become important again. The Flemish Government encourages farmers to cultivate clovers (*Trifolium pratense* L., and *Trifolium repens* L.), lucerne (*Medicago sativa* L.) and grass/clover to produce more farm-grown proteins and to decrease the use of mineral fertilizers. In this study the effect of N fertilization on the botanical composition, dry matter (DM) yield, nutritive value and nitrate residue in the soil is investigated.

## Materials and methods

The experiment was carried out in Merelbeke (sandy loam soil, pH 6.1, altitude 11 m, average annual temperature 9.9°C, average annual precipitation 780 mm). Monocultures of red clover (Tp) and white clover (Tr), lucerne (Ms), perennial ryegrass (*Lolium perenne* L., Lp) and Italian ryegrass (*Lolium multiflorum* Lam., Lm) and 7 grass/legume mixtures were established as a complete block design with 3 replicates in April 2003 at recommended seeding rates. The proportion of Ms, Tp and Tr in the grass/legume mixtures was 40, 30 and 10% respectively. Three levels of chemical N fertilization were established for all the treatments: 0, 105 and 265 kg N ha<sup>-1</sup> y<sup>-1</sup>, hereafter designated N0, N105 and N265. The highest N-level corresponded to the maximum level of active N that can be applied in accordance with the Flemish manure action plan (MAP III). Nitrogen of N105 was applied for the first (52.5 N) and the second cut (52.5 N). Nitrogen of N265 was divided over the first 3 cuts: 90-90-85 kg N ha<sup>-1</sup> y<sup>-1</sup>. All treatments received 60 kg P<sub>2</sub>O<sub>5</sub> and 360 K<sub>2</sub>O ha<sup>-1</sup> y<sup>-1</sup>. The plots (12 m<sup>2</sup> gross area, 8.4 m<sup>2</sup> net area) were cut 3 times in the first year and 5 times in the following years. At each cut dry matter (DM) yield was measured and a grab subsample was separated into the individual sown species and an unsown part. Samples were analysed by NIRS to determine chemical composition and



digestibility and energy (fodder unit milk-VEM) and protein content (true protein digested in the small intestine – DVE and rumen degraded protein balance – OEB) were calculated. Soil nitrate-N (in 0-90 cm) was determined yearly at beginning of November.

## Results and discussion

The proportion of weeds (= unsown species) in the total DM yield in the period 2004-2006 was very low for grass/legume mixtures (< 2%) and Tp (2%), intermediate for Ms (5%) and Lm (6%) and relatively high for Tr (8%) and Lp (11%). In the monoculture of the ryegrasses white clover was the main intruder and in the monocultures of the legumes *Lolium* species and *Poa annua* L. were the main invasive species. In the monocultures of grasses weeds significantly ( $P < 0.01$ ) decreased with fertilization while weeds tended ( $P = 0.008 - P = 0.08$ ) to increase with fertilization in the legume monocultures. There was a tendency of a decrease in invasion by white clover when more N was used in the grass treatments.

The use of N had no influence on the legume content in the monocultures of Tp, Tr, Ms and the effect on DM yield was only significant ( $P = 0.04$ ) for the N265 treatment of Ms. N105 and N265 resulted respectively in an additional yield of 0.42 and 0.82 Mg DM ha<sup>-1</sup> or 4.0 and 3.1 kg DM kg<sup>-1</sup> fertilizer N in comparison with N0 (Table 1).

Table 1. Effect of N fertilization on botanical composition and dry matter yield of grass/legume mixtures, legumes and grasses (average of 2004-2006; Ms: *Medicago sativa*; Tp: *Trifolium pratense*; Tr: *Trifolium repens*; Lm: *Lolium multiflorum*; Lp: *Lolium perenne*).

Treatment	Content of sown legumes % in Dry Matter (DM)			DM yield Mg ha <sup>-1</sup> year <sup>-1</sup>			DM yield kg kg <sup>-1</sup> fertilizer N	
	N0	N105	N265	N0	N105	N265	N105	N265
Tp + Lm	67	51	30	13.23	14.39	14.45	11.0	4.6
Tp + Lp	73	68	56	13.20	13.97	14.02	7.4	3.1
Tp + Tr + Lp	61 + 15	65 + 6	43 + 14	12.65	13.87	14.60	11.6	7.4
Tr + Lp	57	50	30	11.59	12.60	13.20	9.6	6.1
Ms + Lm	81	65	56	15.10	16.48	17.05	13.1	7.3
Ms + Lp	87	82	80	15.91	16.34	16.38	4.1	1.8
Ms + Tr + Lp	85 + 6	79 + 5	68 + 11	15.45	16.64	16.64	11.4	4.5
mean								
grasses				5.50	9.32	11.23	36.4	21.6
legumes	96	94	94	12.31	12.73	13.13	4.0	3.1
grass/legumes	76	68	55	13.88	14.90	15.19	9.7	5.0

The proportion of legumes in the grass/legume mixtures significantly ( $P < 0.02$ ) decreased when more N was applied but the level of legumes was substantial, especially for the combinations with Lp in N105. For grass/legume mixtures, N105 and N265 resulted in an increase of the DM yield by 1.02 and 1.31 Mg ha<sup>-1</sup> respectively. It is obvious that the small difference in DM yield between these two N levels (0.29 Mg ha<sup>-1</sup>) did not economically justify the use of 265 kg N on grass/legume mixtures. In comparison with N0 the use of 105 kg N ha<sup>-1</sup> resulted in an additional yield of 9.7 kg DM kg<sup>-1</sup>N which corresponded with an cost price of 0.12 € kg<sup>-1</sup> DM (765€ Mg<sup>-1</sup> N for fertilizer and 20€ fertilizer application<sup>-1</sup> ha<sup>-1</sup>). The growth of Lp and Lm is still limited by a N fertilization of 265 kg N ha<sup>-1</sup> (Behaeghe and Carlier, 1974; Table 1) but the MAP III regulation does not allow to apply more N.

In this experiment no consistent correlation was found between the energy content of the dry matter and the level of N fertilization (Table 2). Tr, followed by Lp, Lm and Tr + Lp had the highest energy content (> 800VEM) and Ms (713VEM) as well as grass/lucerne mixtures (728-756 VEM) the lowest. One can expect that a higher proportion of grasses in the mixture, caused by a higher N use, should increase the energy content of the dry matter but this was not observed in this experiment. The protein content, in terms of DVE and OEB, was more

affected by type of monoculture or mixture than applied N-level. The effect of N fertilization was significant ( $P < 0.01$ ) but not systematic for every monoculture/mixture although the protein content was mostly the largest when no N is applied. The pure stands of the legumes Tr and Tp had significant (Tukey test,  $\alpha = 0.05$ ) larger levels of protein in the DM (except Tp vs Tr + Lp for DVE and vs Ms + Lp( + Tr) for OEB). The monocultures of ryegrasses, handicapped by the inferior level of N fertilization, had significant lower protein levels than the mixtures. The average DVE content of the grass/legume mixtures was  $84 \text{ g kg}^{-1} \text{ DM}$ , which is comparable to perennial ryegrass in favourable growing conditions. The mixtures with Lm had a significant smaller DVE protein content in comparison with the mixtures with Lp. This is in accordance with previous observations (De Vliegheer and Carlier, 2005). To avoid leaching, the nitrate nitrogen content in the soil profile 0-90 cm should not exceed  $90 \text{ kg ha}^{-1}$  at the end of the growing season. The nitrate residue in the soil was relatively higher for the monocultures of legumes ( $17\text{-}22 \text{ kg N ha}^{-1}$ ) in comparison with grass/legume mixtures ( $10\text{-}15 \text{ kg N ha}^{-1}$ ) and grasses ( $6\text{-}8 \text{ kg N ha}^{-1}$ ) but they were always far below the legal limit of  $90 \text{ kg ha}^{-1}$  (Table 2). There was no effect of N fertilization on the nitrate residue in the soil.

Table 2. Effect of N fertilization on forage quality of grass/legume mixtures, legumes, grasses and nitrate residue in the soil (average of 2004-2006; Ms: *Medicago sativa*; Tp: *Trifolium pratense*; Tr: *Trifolium repens*; Lm: *Lolium multiflorum*; Lp: *Lolium perenne*).

Treatment	Energy content			Protein content						NO <sub>3</sub> -N in the soil		
	VEM kg <sup>-1</sup> DM			DVE g kg <sup>-1</sup> DM			OEB g kg <sup>-1</sup> DM			kg ha <sup>-1</sup>		
	N0	N105	N265	N0	N105	N265	N0	N105	N265	N0	N105	N265
Tp + Lm	775	788	795	83	77	76	43	23	22			
Tp + Lp	760	773	780	85	86	84	49	50	46	8	19	16
Tp + Tr + Lp	784	787	787	89	87	85	49	48	46			
Tr + Lp	843	829	826	92	87	84	42	36	30	9	14	13
Ms + Lm	733	756	746	81	79	78	49	40	41			
Ms + Lp	728	740	736	84	82	84	56	48	54	12	12	14
Ms + Tr + Lp	755	753	746	89	87	85	58	54	56			
mean												
grasses	806	842	824	71	70	71	-9	-9	-2	6	8	8
legumes	776	777	781	95	94	95	63	60	61	17	17	22
grass/legumes	768	775	774	86	84	82	49	43	42	10	15	14

VEM: fodder unit milk; DVE: true protein digested in the small intestine; OEB: rumen degraded protein balance.

## Conclusions

Mineral N fertilization up to  $265 \text{ kg N ha}^{-1}$  had no influence on legume content or on DM yield (except the N265 treatment of lucerne) in monocultures of clover or lucerne. The proportion of legumes in the grass/legume mixtures decreased when more N was used but the level of legumes remained substantial ( $> 50\%$  of the DM). For the mixtures, the use of 105 or  $265 \text{ kg N ha}^{-1} \text{ y}^{-1}$  resulted in an additional yield of about 1.0 and  $1.3 \text{ Mg ha}^{-1}$ , respectively. No relationship was found between the energy content and the level of N fertilization. The protein content was more affected by type of seed mixture than by applied N-level. Growing legumes or grass/legume mixtures under a cutting regime gave no difficulties in terms of nitrate residue.

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# Genotypic and symbiotic characters among rhizobia nodulating red clover in soils of northern Scandinavia

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## Abstract

Indigenous populations of *Rhizobium leguminosarum* bv. *trifolii* nodulating red clover (*Trifolium pratense* L.) were sampled from two field sites, in northern Sweden and northern Norway, at different times of the growing season. A total of 431 nodule isolates were characterized genetically by targeting both chromosomal and symbiotic genes. The chromosomal analysis revealed considerable genetic variation within the isolated populations that was more influenced by geographical origin than sampling time. Restriction fragment length polymorphism analysis of the *nodEF* gene on the symbiotic plasmid revealed a high proportion of *nod* genotypes common to the two sites. The symbiotic efficiency of eight isolates, representing both dominating and rare *nod* genotypes, was measured in a greenhouse experiment using the <sup>15</sup>N isotope dilution method. All tested isolates except one, representing a very rare genotype, showed high N<sub>2</sub> fixation rates in symbiosis with the host plant. There was thus no correlation between relative abundance and symbiotic efficiency among the sampled rhizobial genotypes, and we conclude that effective N<sub>2</sub>-fixing strains of *Rhizobium leguminosarum* bv. *trifolii* nodulating red clover are common and genetically diverse in these northern Scandinavia soils.

Keywords: clover, diversity, genetic fingerprinting, *Rhizobium leguminosarum* bv. *trifolii*, symbiotic efficiency

## Introduction

Red clover (*Trifolium pratense* L.) is the most widely used forage legume in Scandinavia (Frame *et al.*, 1998). Spontaneous nodulation of red clover by *Rhizobium leguminosarum* bv. *trifolii* present in the soil is frequently observed when it is grown in Scandinavia, and it is therefore not common practice to use rhizobial inoculants for red clover. Indigenous soil populations of rhizobia nodulating clovers are often highly genetically diverse, and typically consist of one or a few dominating genotypes and several genotypes present at lower frequency (Leung *et al.*, 1994; Zézé *et al.*, 2001; Fagerli and Svenning, 2005). However, there is only limited information available regarding the symbiotic efficiency of indigenous red clover-nodulating rhizobia. It is also not known to what extent dominating rhizobial genotypes have higher symbiotic N<sub>2</sub> fixation efficiency compared to rare genotypes. With the aim to investigate the potential relationship between relative abundance in a field population and N<sub>2</sub> fixation efficiency, the genotypic and phenotypic composition of rhizobial populations isolated from field-grown red clover trap plants were analysed at two geographical sites: northern Norway (Tromsø) and northern Sweden (Umeå).

## Materials and methods

In total 431 *R. l.* bv. *trifolii* isolates were sampled from trap plants of red clover (*Trifolium pratense* L.) cv. Betty nodulated in agricultural fields in Tromsø (Norway, (69°40'N, 18°56'E) and Umeå (Sweden 63°45'N, 20°17'E) during summer and autumn. The distance



between the two sampling sites is about 700 km. The genetic variation among sampled isolates was analysed based on variations within their chromosomal as well as their symbiosis (sym) plasmid genomes. DNA fingerprinting by PCR with Enterobacterial Repetitive Intergenic Consensus (ERIC) primers was used to characterize the chromosomal diversity (de Bruijn, 1992), and the portion of the genome associated with symbiotic characters was characterized by RFLP analysis of the *nodEF* gene on the symbiotic plasmid (Fagerli and Svenning, 2005). The symbiotic efficiency of eight isolates, representing both dominating and rare *nod* types, was measured in a greenhouse experiment using the  $^{15}\text{N}$  isotope dilution method. Statistically significant differences in  $\text{N}_2$  fixation efficiency were tested by the two-sample Student's *t*-test using the Minitab 14 statistical software (Minitab inc., State College Pennsylvania, PA, USA, 2003). The experimental procedures are described in detail in Duodu *et al.* (2007).

## Results and discussion

The sampled field populations of *R. l. bv. trifolii* were large, around  $10^4$  infective cells per g dry soil in Tromsø and in the range  $10^4$  to  $10^5$  infective cells per g dry soil in Umeå. The fingerprinting of chromosomal DNA revealed 114 different ERIC types. Of these, 62 ERIC types were detected in Tromsø and 53 in Umeå, and only one ERIC type was found at both sites. On the other hand, the *nodEF*-RFLP analysis revealed a high proportion of *nod* types common to the two sites (Figure 1a). In total, 14 different *nodEF* types were found among the 431 isolates. Of these 14 genotypes, three were unique to the populations from Tromsø or Umeå, while eight were common to both geographical sites. Several *nodEF* types were represented by more than one ERIC type, as indicated by the numbers on top of the bars in Figure 1a. Interestingly, there were also several examples of isolates with the same chromosomal (ERIC) identity that differed in their symbiotic (*nodEF*) identities (Duodu *et al.*, 2007). These findings provide evidence for exchange or transfer of *sym* plasmids among red clover-nodulating rhizobia.

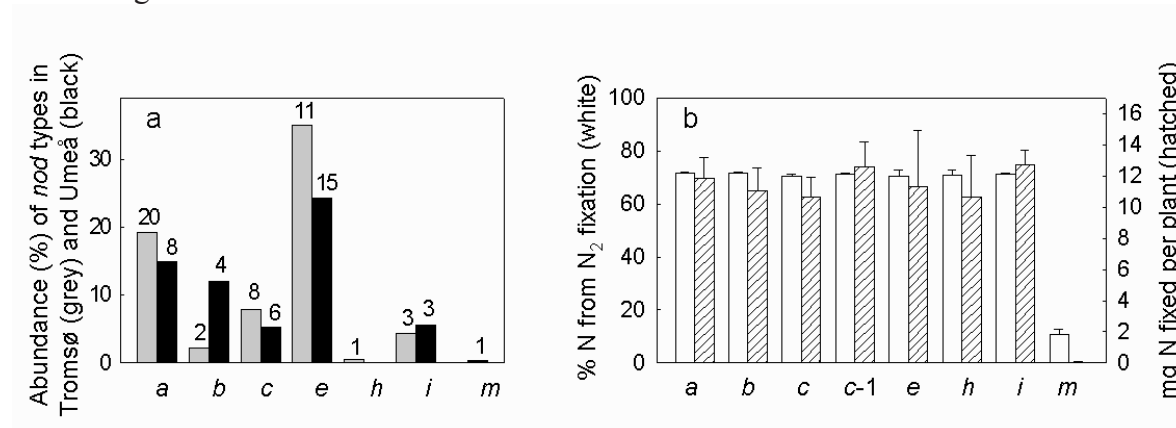


Figure 1. Relative abundance of seven *nod* types at the two sampling sites (a) and  $\text{N}_2$  fixation in red clover plants inoculated with *R. l. bv. trifolii* isolates representing different *nod* types (b). Numbers on top of the bars (a) show the number of ERIC types represented by each *nod* type.  $\text{N}_2$  fixation data are mean values + SD,  $n = 5$ . *Nod* types *c* and *c-1* are identical, but represent different ERIC types.

Among the eight isolates that were tested for their symbiotic efficiency, all except one showed high  $\text{N}_2$  fixation (Figure 1b). Apart from the large difference between *nodEF* type *m* and all other *nodEF* types, the only statistically significant ( $P < 0.05$ ) difference in  $\text{N}_2$  fixation was that *nodEF* type *c-1* fixed more  $\text{N}_2$  than *nodEF* type *c*. The isolate with the *nodEF* type *m*,

showing a very low N<sub>2</sub> fixation rate, represented a very rare genotype – its ERIC type was found in only one of the 431 isolates. Apart from this extremely rare genotype, there was no relationship between the relative abundance of different rhizobial genotypes in the sampled populations and their N<sub>2</sub> fixation efficiency in symbiosis with the host plant. The ability of red clover to form efficient symbiosis with nearly all genotypes recovered at different times of the growing season highlights the agronomic value of this legume in northern Scandinavia, including subarctic regions.

## Conclusions

Effective strains of *R. l. bv. trifolii* nodulating red clover were abundant in the sampled soils, and despite the distance between Tromsø and Umeå several effective genotypes were common to both sites. There was no correlation between relative abundance of rhizobial genotypes and their symbiotic efficiency. The sampled soils harboured large rhizobial populations and the abundance of inefficient genotypes was very low. There is thus no need to inoculate red clover with *R. l. bv. trifolii* when cultivating this species in northern Scandinavia.

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# Genetic characterization of indigenous root nodule bacteria from *Trifolium alexandrinum* L. cultivated on the island of Sardinia

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## Abstract

In order to provide information about a natural population of root nodule bacteria with respect to a productive Italian annual forage legume *Trifolium alexandrinum* L. (berseem clover), the genotypic characterization of the rhizobia was determined by polymerase chain reaction (PCR) using repetitive extragenic palindromic (REP) sequences. The aim of the research was to identify the biotypes present in the root nodules of plants grown in the field belonging to two varieties (Marmilla and Sacromonte), and to characterize them by some phenotypic traits. Ten plants per variety and five nodules per plant were collected. Scoring of the nodules, plant length, root length and plant dry weight (DW) were recorded. A total of 10 different biotypes out of 43 isolates were detected. Seven biotypes were common to the varieties with the dominance of biotype b. No significant differences in the plant phenotypic traits and forage yield were found.

Keywords: berseem clover, REP-PCR, genetic diversity, rhizobia

## Introduction

Symbiotic relationship established between the legumes and the rhizobia present in the soil is known to be a fundamental process for plant growth and crop productivity. On the island of Sardinia, farmers do not generally inoculate because the widespread occurrence of native forage legumes is an index of the presence of indigenous rhizobia. Berseem clover (*Trifolium alexandrinum* L.) is an annual forage legume well adapted to the Mediterranean climate, characterized by a good productivity and a high nutritive value for animal feeding. The aim of the research was to characterize genetically the root nodule bacteria from two Italian varieties (Marmilla and Sacromonte) grown in the field by repetitive extragenic palindromic (REP) PCR and to study some phenotypic traits of the plants.

## Materials and methods

The trials were carried out during 2006-2007 on the Bonassai Experimental Farm (40°39'46''N, 8°21'46''E; 33m a.s.l.) in the North-West of Sardinia, on a flat clay-loam calcareous soil (pH 7.5). The climate is Mediterranean with an average annual rainfall of 589 mm. Two varieties (Marmilla and Sacromonte) of *T. alexandrinum*, belonging to a wider experiment, were sown in autumn in 2 plots of 12 m<sup>2</sup> per variety with a seed rate of 3g m<sup>-2</sup>. Forage yield was determined by cutting two 0.5 m<sup>2</sup> samples of each plot in February, March and May. After 12 weeks from sowing, 10 plants of each variety were collected randomly. Each plant was split into the aerial part and the root, and their lengths were measured. Some phenotypic traits such as plant length, root length and plant dry weight, were recorded. Roots were scored for nodule number, form and colour. Five nodules per plant were randomly surface-sterilized by sequential washings with 70% (v/v) ethanol for 1-2 minutes, sodium hypochlorite for 3 minutes and six rinses in sterile distilled water (SDW). The individual nodules were crushed aseptically in a drop of SDW by sterilized forceps and the suspension was streaked on yeast extract mannitol agar (YMA) plates containing 0.0025% (w/v) Congo Red and put in a 28 °C incubator. Isolations were performed on three-day-old colonies. Root

nodule isolates were purified by repeated streakings of single colonies on the plates containing yeast extract mannitol agar (YMA), frozen at -80 °C and reactivated on YMA at 28 °C for DNA extraction and PCR analyses. Bacterial cell preparation for REP-PCR amplification was carried out on 43 isolates as described by Laguerre *et al.* (1997). Oligonucleotide primer sequences (Primm) and PCR conditions were as indicated by Versalovic *et al.* (1991) and de Bruijn (1992), respectively. Amplification products were separated by horizontal electrophoresis on 1% (w/v) agarose gel (Sigma) in Tris-acetate EDTA at 90 V for 2 h. Averages were processed by t-test.

## Results and discussion

REP-PCR allowed us to identify at strain level 43 root nodule isolates from berseem clover. The different REP-PCR profiles from the root nodule isolates produced a range of bands from 200 to 3,500 base pairs (bp) in length, and the number of bands per strain varied between 3 and 9 (Figure 1).

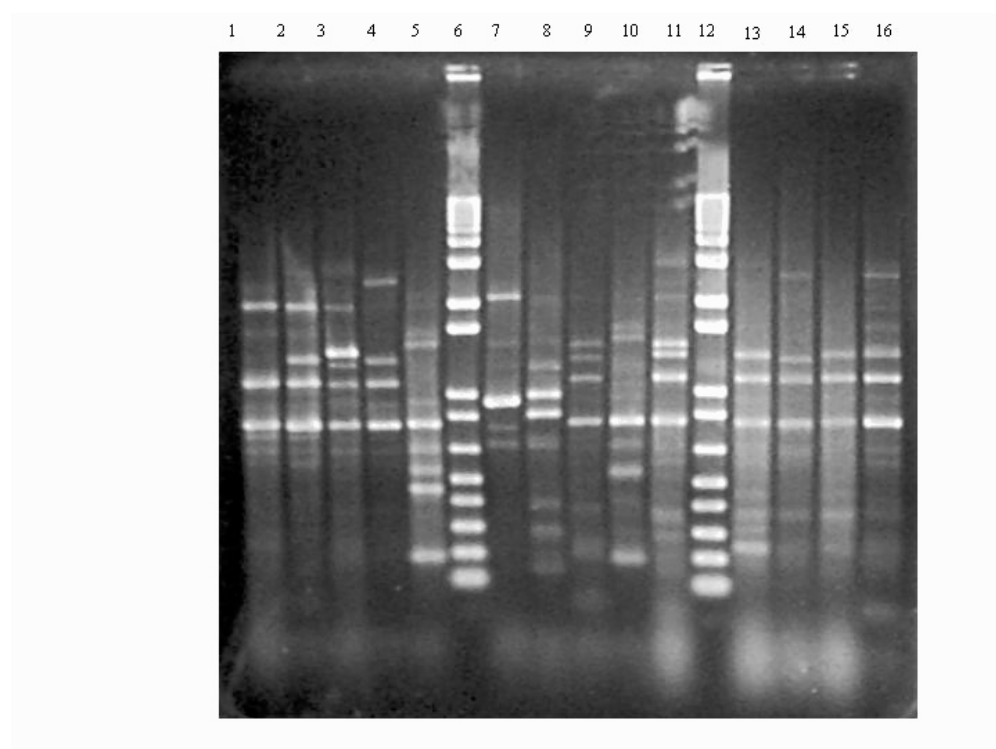


Figure 1. REP-PCR profiles from root nodule isolates from *Trifolium alexandrinum* L. Marmilla variety: lanes 1-5, 7, 8 (biotypes: a, g, c, e, d, f, i); Sacromonte variety: lanes 9-11, 13-16 (biotypes: c, d, c, b, e, b, e); lanes 6-12 Marker: 1 kb Plus DNA Ladder (Invitrogen) (100-12,000 bp).

The biotypes obtained by REP-PCR showed a remarkable biodiversity: in fact 10 different biotypes (a, b, c, d, e, f, g, h, i, l) out of 23 isolates (43.5%) were found in the roots of *T. alexandrinum* var. Marmilla and 7 different biotypes (a, b, c, d, e, f and l) were observed among the 20 isolates (35%) from Sacromonte (Table 1). Seven biotypes were common to the varieties with the dominance of biotype b. As far as the number of different biotypes inside a single plant is concerned, we found up to 4 different biotypes colonizing the same root (data not shown). This variability is quite interesting and suggests some possible hypotheses: a) different strains of the same species of rhizobia could be present in the roots of *T. alexandrinum*; b) diverse bacteria represented by different genera and species are associated with root nodules. Various studies reported the presence of heterogeneous rhizobial

isolates from pasture legumes native to Sardinia (Safranova *et al.*, 2007) and diverse genera of bacteria associated with root nodules of spontaneous legumes in Mediterranean soils (Zakhia *et al.*, 2006).

There were no significant differences between the two varieties in annual forage yield, plant length, root length and plant dry weight (Table 1). However, the number of nodules differed significantly in Marmilla and Sacromonte, showing greater values in the first variety ( $P < 0.05$ ).

Table 1. Annual forage yield (DM), plant length, root length, nodule number, dry weight (DW) and REP types frequency in two cultivars of *T. alexandrinum* L.

	DM	Plant length	Root length	Nodule	DW	REP types frequencies
Varieties	t ha <sup>-1</sup>	cm	cm	n°plant <sup>-1</sup>	mg plant <sup>-1</sup>	
Marmilla	6.4 ± 12.6	29.9 ± 3.1	11.9 ± 1.7	39 ± 14	200 ± 94.3	a(3);b(7);c(4);d(3); e(1);f(1);g(1);h(1); i(1); l(1).
Sacromonte	6.6 ± 12.3	31.9 ± 2.5	10.9 ± 3.0	27 ± 6	170 ± 105.9	a(1);b(6);c(5);d(4); e (2); f(1); l(1).
<i>P</i>	ns	ns	ns	*	ns	

\*data differ significantly;  $P < 0.05$  (t-test); ns: not significant.

Generally, the nodules observed were oblong-shaped, white-pink in colour and of small size (0.5 mm) or fan-shaped and of medium size (2.5-3 mm).

## Conclusions

This study is the first step in the genetic characterization of indigenous root nodule bacteria from *T. alexandrinum* L. cultivated on the island of Sardinia. Additional primer sets are to be used to genotype the isolates and sequencing of 16S rDNA will allow us to identify the isolates at species level.

## Acknowledgments

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# Comparison of yields and biological nitrogen fixation of two legumes grown in different silt soils

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## Abstract

The legume species usually sown in mixtures with grasses do not grow very well in silt soils. Biological characteristics of birdsfoot trefoil (*Lotus corniculatus*) and alsike clover (*Trifolium hybridum*) are still poorly documented, though they can resist drought and/or water excess. We compared, (i) early development, and (ii) biological nitrogen fixation by four varieties of each species grown in silt soils. Four varieties of birdsfoot trefoil and of alsike clover were sown without inoculation in 4.5 l pots filled with five different silt soils ( $n = 4$ ). At the end of the experiment, above-ground plant parts were counted, measured, dried, weighed and prepared for  $^{15}\text{N}$ : $^{14}\text{N}$  measurements. Within each species, the number of leaves and ramifications, leaf area, and dry weights did not differ between varieties. Conversely, results differed markedly between soil types. Nodules were found in all pots, but there were significant differences in the amount of fixed nitrogen among soils in birdsfoot trefoil as well as in alsike clover: the proportion of the plant N derived from fixation varied from 0 to about 80%.

Keywords:  $\text{N}_2$  biological fixation, *Lotus corniculatus*, *Trifolium hybridum*, silt soil, grassland legumes

## Introduction

The use of grass/legume mixtures in grasslands is known to be of interest for reducing application of industrial N fertilizers. Høgh-Jensen and Schoering (2001) reported that as much as 92% of the N rhizo-deposited in a clover mixture could be derived from biological nitrogen fixation (Nd<sub>fa</sub>). Most of the legume species usually sown with grasses in mixtures are not well adapted to poor soils, such as silt soils commonly encountered in the west of France (more than 30% silt). These soils tend to form compact layers below the surface, restricting water and air movements, with consequences on root development and  $\text{N}_2$  fixation. Birdsfoot trefoil (*Lotus corniculatus* L.) and alsike clover (*Trifolium hybridum* L.) are still poorly documented, though they seem adapted to poor agronomic conditions (Seguin *et al.* 2000). The aims of this study were to compare (i) early development, and (ii) the amount of  $\text{N}_2$  fixed by several varieties of both species grown in different kinds of silt soils.

## Materials and methods

Four varieties of birdsfoot trefoil (cv. 'Albena', 'Leo', 'Gran San Gabriele' and 'Lotanova') and of alsike clover (cv. 'Dawn', 'Dixon', 'Buffalo' and 'Ermo') were sown separately in 4.5 l pots filled with five different loams (28-50% silt, < 52% sand) or silt loams (> 50% silt, < 52% sand) collected from the 20 cm upper layer of harvested fields, at five locations in the west of France. Soils were roughly blended and stones were not removed; one sample of each soil was analysed. Properties measured were textural characteristics, total C and N, mineral N, proportion of organic matter, pH, Olsen phosphate, cation exchange capacity Metson (exchangeable  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ ,  $\text{K}^{+}$ , and  $\text{Na}^{+}$ ). For each variety and each soil, 12 seeds were carefully distributed around a central circle drawn on the wet soil surface (4 replicates). To provide a control for assessing Nd<sub>fa</sub> by the  $^{15}\text{N}$  natural abundance method (Hansen and Vinther, 2001), a mixture of 50% rye-grass/50% red fescue was grown in the same conditions

as the legumes. The 180 pots were distributed at random on culture tables in a greenhouse (mean temperature 24.3°C; mean hygrometry 67.4%). Throughout the experiment, they were simultaneously watered by sub-irrigation. After the development of the first true leaf, seedlings were thinned to six plants per pot. Plants were harvested after nine weeks. For birdsfoot trefoil, the number of leaves and stems per plant were counted. For alsike clover, the number of leaves, the plant height and the total leaf area of each plant (planimeter) were recorded. Roots were examined. Above ground plant parts from each pot were pooled and dried at 60 °C before weighing. They were prepared for  $^{15}\text{N}$ : $^{14}\text{N}$  measurements (INRA of Laon, France). The proportion of the legume-N derived from fixation (%Nd<sub>fa</sub>) was calculated according to Hansen and Vinther (2001). For each variety within a species, we compared the results obtained with different soils. Subsequent analyses were carried out, i) after pooling results obtained with all varieties of a each species grown on the same soil, or ii) after pooling results obtained on all soils for a particular variety.

### Results and discussion

Soils A and B were very similar in terms of their textural characteristics, while they differed in their organic matter contents and chemical characteristics (Table 1). Both contained about 50% sand and 10% clay. C, D and E contained more than 45% silt and less than 35% sand. Soil E (66% silt and 15% sand) differed markedly from C and D (about 48% silt and 32% sand).

Table 1. Main characteristics of the studied soils.

Soil	A	B	C	D	E
% Clay	10	10.4	19.4	13.3	15.9
% Silt	39.5	35.6	47.9	49.4	66.5
% Sand	47.8	50	30.7	34.5	15.6
% OM <sup>a</sup>	2.5	4	1.9	2.8	2
CEC <sup>b</sup>	6.2	13.2	8.3	10	9.8
pH <sub>KCl</sub> (pH <sub>H2O</sub> )	5.0 (5.8)	5.3 (6.2)	5.9 (6.6)	4.7 (5.8)	5.2 (6.1)
N (mg kg <sup>-1</sup> ) <sup>c</sup>	0.7	1.4	0.8	1.1	0.7
P (mg kg <sup>-1</sup> )	53.6	21.3	37.9	40.5	20.5
K (mg kg <sup>-1</sup> )	166	141.1	572	273.9	107.9
Mg (mg kg <sup>-1</sup> )	60.3	72.4	108.5	102.5	78.3
Ca (mg kg <sup>-1</sup> )	786.5	1708	1101	1058.2	1780.3

<sup>a</sup>organic matter <sup>b</sup>Cation Exchange Capacity Metson (mEq 100 g<sup>-1</sup>) <sup>c</sup>Mineral N.

In birdsfoot trefoil, the plants with the greatest dry weights also had the greatest number of leaves and stems. In alsike clover the plants that were tallest had the greatest number of leaves and the largest leaves. Within each species, dry weights and N concentrations did not differ between varieties. Conversely, results differed markedly between soils: for the two legume species, the greatest yields and plant-N concentrations were obtained in D, and the lowest in E (Table 2). D had high levels of mineral N and exchangeable P, K, Mg, but a low level of Ca, while E had low levels of N, P, Mg, but high levels of Ca, and more than 50% silt. For a given soil, dry weight of above ground parts did not differ between species. Nodules were observed in all pots, but they seemed to be less abundant for plants grown in B and E. Morphology and size of nodules differed between soils, species and varieties. %Nd<sub>fa</sub> was nil for the two species in soil B (Table 2). As plants were harvested during early growth, this result may be due either to a delay in nodule formation and activity in B compared to the other soil (Williams 1988) or to a lack of plant-*Rhizobium* compatibility. In E, results depended on the variety: in birdsfoot trefoil, %Nd<sub>fa</sub> was nil for Lotanova and Gran San Gabriele, but reached more than 50% of plant-N with Albena and Leo. In alsike clover, %Nd<sub>fa</sub> was 44% for Ermo, 19% for Dixon, 10% for Dawn and only 1% for Buffalo. For the two species, %Nd<sub>fa</sub> was

found to be negatively correlated with inorganic N of the soil, Ca, and the percentage of organic matter (Spearman  $r$ ;  $P < 0.0001$ ), but positively correlated with P and K ( $P < 0.0001$ ). Additionally, with birdsfoot trefoil, %Ndfa was negatively correlated with the pH of the soil ( $r = -0.375$ ;  $P = 0.018$ ) and with the percentage of clay ( $r = -0.408$ ;  $P = 0.0006$ ). These relationships help to explain why symbiotic nitrogen fixation was more efficient (or started earlier) in A, C and D, especially for alsike clover in C and D.

Table 2. Mean dry weights, N concentration of above ground parts, and part of the plant-N derived from fixation (%Ndfa) in birdsfoot trefoil, alsike clover and reference plants.

Soil	A	B	C	D	E	<i>p</i>
<i>Lotus corniculatus</i>						
Dry weight per plant (g)	0.9 <sup>bc</sup>	0.9 <sup>b</sup>	0.7 <sup>c</sup>	1.3 <sup>a</sup>	0.5 <sup>d</sup>	***
N concentration (g kg <sup>-1</sup> )	18.1 <sup>b</sup>	20.5 <sup>b</sup>	11.9 <sup>c</sup>	26.1 <sup>a</sup>	13.6 <sup>c</sup>	***
Ndfa (%)	73.4 <sup>a</sup>	0.4 <sup>c</sup>	47.8 <sup>b</sup>	41.3 <sup>bc</sup>	19.4 <sup>c</sup>	***
<i>Trifolium hybridum</i>						
Dry weight per plant (g)	0.7 <sup>c</sup>	0.6 <sup>d</sup>	0.8 <sup>b</sup>	1.1 <sup>a</sup>	0.5 <sup>e</sup>	***
N concentration (g kg <sup>-1</sup> )	21.0 <sup>b</sup>	17.8 <sup>b</sup>	25.4 <sup>b</sup>	32.9 <sup>a</sup>	14.2 <sup>c</sup>	***
Ndfa (%)	65.3 <sup>b</sup>	0	80.7 <sup>a</sup>	54.3 <sup>b</sup>	22.2 <sup>c</sup>	***
<i>Reference plants</i>						
Dry weight (g)	4.3 <sup>cd</sup>	8.0 <sup>a</sup>	5.9 <sup>b</sup>	5.2 <sup>bd</sup>	4.3 <sup>c</sup>	**
N concentration (g kg <sup>-1</sup> )	13.0 <sup>bc</sup>	11.8 <sup>bc</sup>	11.6 <sup>b</sup>	13.8 <sup>ac</sup>	14.4 <sup>ab</sup>	*

Different letters indicate significant difference between soils.

Results of 4 varieties for each legume species ( $n = 16$ ) \* $P < 0.05$ , \*\* $P < 0.005$ ; \*\*\* $P < 0.0001$ .

## Conclusions

In the acid loam with the highest extractable P, Ndfa during early plant growth was about 70% of the plant-N for birdsfoot trefoil and alsike clover. On such soils, it would be interesting to use both species in mixture with grasses. At this growth stage, in soils with 50% silt and 30% sand, biological nitrogen fixation was more efficient in alsike clover than in birdsfoot trefoil: in these soils, alsike clover would be a better choice than birdsfoot trefoil for grass/legume mixtures, since its earlier start of N<sub>2</sub> fixation activity gives the legume a clear advantage in competition with grasses.

## Acknowledgements

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## Analysis of *Festulolium* and hybrid ryegrass (*Lolium x boucheanum*) dry matter yield stability

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### Abstract

Under Latvian climatic conditions, forage grasses are the main source of fodder for breeding cattle. *Festulolium* hybrids are among the most persistent and productive of the grasses used in many European countries, especially in adverse environments. Field trials were established on a sod-podzolic soil and fertilized with N 120 (60 + 60), N180 (60 + 60 + 60), P 78 and K 90 kg ha<sup>-1</sup>. Forages were harvested three times during the growing season. On the basis of the experiments in the years 2003-2006, significant differences in dry matter (DM) yield and winter hardiness were found between the first, second and third years. Differences between varieties in DM yield were highly significant and showed a similar tendency during all three years. Analysis of yield distribution between the three cuts showed that year of sward use had very great effect on DM yield of the first cut. The average DM yields of *Festulolium* cultivars were 3.99 Mg ha<sup>-1</sup> greater, and those of hybrid ryegrass were 2.37 Mg ha<sup>-1</sup> greater, when compared with perennial ryegrass.

Keywords: *Festulolium*, *Lolium hybridum*, productivity, regrowth

### Introduction

Many dairy husbandry producers in Latvia are shifting towards intensive systems of production. The level of the productivity and stability mostly depends on the genetic potential of the forage grass species. *Lolium* species (considered the ideal grasses for European agriculture) are not sufficiently robust in less favoured areas. In the Baltic region hybrid ryegrasses are not widely used because of their unsatisfactory response to cold conditions and decline of productivity over successive years (Gutmane and Adamovich, 2006). Introduction of biotic and abiotic stress tolerance from *Festuca* spp. into *Lolium* spp. has been made for many years, thus offering unique opportunities for the production of versatile hybrid varieties with new combinations of useful characters suited to modern grassland farming (Humphreys *et al.*, 2006). An important requirement for *Festulolium* is combining of ryegrass characters, such as productivity, growth potential and feeding quality, with those from fescues: stress resistance in wintering and resistance to drought during the growth period (Casler *et al.*, 2002).

The objective of this research was to investigate dry matter (DM) yield formation under cutting regime, and sward persistency of *Festulolium* and hybrid ryegrass (*Lolium x boucheanum*) varieties, under the agro-ecological conditions of Latvia.

### Materials and methods

Field trials were conducted in Latvia on sod-podzolic soils. The pH was 7.1 and phosphorus and potassium levels were 253 and 198 mg kg<sup>-1</sup> respectively, and organic matter content was 31 g kg<sup>-1</sup>. Swards were composed of: perennial ryegrass 'Spidola' (control); *Festulolium* 'Perun' (*L. multiflorum* x *F. pratensis*), 'Punia' (*L. multiflorum* x *F. pratensis*), 'Saikava' (*L. perenne* x *F. pratensis*), 'Lofa' (*L. multiflorum* x *F. arundinacea*), 'Felina' (*L. multiflorum* x *F. arundinacea*), 'Hykor' (*L. multiflorum* x *F. arundinacea*); and hybrid ryegrass 'Tapirus'

(*L. multiflorum* x *L. perenne*). The trial was sown in May 2002 and 2003, without cover crop, at a sowing rate of 1,000 germinating seeds m<sup>-2</sup>. Swards were cut three times per season. The experimental data was subjected to analysis of variance (ANOVA).

## Results and discussion

A higher herbage production rate from newly established grass swards is one of the expectations in temporary grasslands. Longevity of *Festulolium* swards was affected by different reasons, such as suitability of each variety to specific conditions, different stress conditions, and the management regime. *Festulolium* hybrids and especially perennial ryegrass often are insufficiently resistant to over-wintering conditions. Meteorological conditions for good grass yields were most favourable in 2004, which had adequate summer rainfall. In years with hot, dry summers (2003, 2005 and 2006) conditions were less favourable for the development of grass plants, and varieties showed differences in the structure of yield formation.

Under cutting conditions the grass yield is often reported to decrease over successive years, with the highest yield in the first harvest year (Hopkins *et al.*, 1990). The sward utilization year has great effect on the DM yield of *Festulolium* hybrids and especially of *Lolium perenne*. Significant DM yield decrease of *Festulolium* hybrids in the second year of sward use in Lithuania was mentioned (Lemeziene *et al.*, 2004). Our results show a substantial decrease in DM yield even between first and second years. The average DM yield distribution during the years of yielding showed significant differences. The maximum yield was obtained in the first year of sward use (Table 1) and it was typical for perennial forage grasses. In the first year of utilization grasses are at the initial stage of their growth and development, and have been exposed only slightly to such negative effects as unfavourable growing or wintering conditions, diseases and inefficient utilization.

Table 1. Average DM yield for three years of sward use (Mg ha<sup>-1</sup>).

Variety		1st year of sward use			2nd year of sward use			3rd year of sward use		
		1st cut	2nd cut	3rd cut	1st cut	2nd cut	3rd cut	1st cut	2nd cut	3rd cut
Spidola	Lp	5.70	1.96	2.97	3.23	1.88	2.09	2.35	0.40	1.90
Tapirus	Lm x Lp	6.88	3.62	3.82	3.47	3.22	2.85	2.86	0.76	2.11
Saikava	Lp x Fp	5.23	4.86	3.61	3.91	1.15	1.77	2.15	0.13	3.35
Lofa	Lm x Fa	7.11	3.77	3.89	3.09	3.58	2.99	2.88	0.92	2.51
Felina	Lm x Fa	10.65	2.07	3.69	4.97	2.93	5.87	5.33	1.60	2.32
Hykor	Lm x Fa	8.63	3.20	4.73	5.19	3.13	5.35	4.78	1.25	4.25
Perun	Lm x Fp	8.19	4.17	4.20	3.89	3.55	3.22	3.50	0.86	2.57
Punia	Lm x Fp	8.78	3.97	3.89	4.46	3.25	3.12	3.74	0.71	2.24
LSD <sub>0.05</sub>		0.58	0.31	0.31	0.42	0.36	0.26	0.33	0.23	0.45

The highest average DM yield in all years of sward use was provided by Lm x Fa cv. 'Hykor' and 'Felina'. These same cultivars had highest plant height before harvesting. These findings correspond with plant morphological characters: cv. 'Hykor' and 'Felina' represent festucoid type of *Festulolium*. During three years of herbage use the lowest yield was provided by perennial ryegrass 'Spidola'. This can be explained by rather poor over-wintering of perennial ryegrass and by plant height. Perennial ryegrass is attributed to short grasses, while the *Festulolium* and hybrid ryegrass to tall grasses. Differences between varieties in DM yield were highly significant and showed a similar tendency during the three years. Compared with Spidola perennial ryegrass, the average DM yield of *Festulolium* cultivars was higher by 3.99 Mg ha<sup>-1</sup>, and of hybrid ryegrass by 2.37 Mg ha<sup>-1</sup>.

A reduction in DM production in the various grasses between the first and third cuts has been reported previously (Tarakanovas *et al.*, 2004). Our results show that *Festulolium* and hybrid

ryegrass swards gave a higher DM yield in the first cut. The influence of the regrowth is very important, especially in less favourable years. The low second cut yield can be explained by hot and dry weather condition during the regrowth period after the first cut.

Results from the analysis of variance showed that in the three years of utilization DM yield for *Festulolium* and ryegrass swards was significantly ( $P < 0.05$ ) dependent on the variety used. A high regrowth factor, and sward utilization-year factor influence on the DM yield was established (Table 2).

Table 2. Analysis of variance for dry matter yields of varieties for three years of sward use.

Source of Variation	SS	Df	MS	F	F <sub>0.05</sub> crit
Variety (V)	119.4	7	17.1	262.24	2.05
Cut (C)	352.2	2	176.1	2707.88	3.04
Year (Y)	345.8	2	172.9	2658.73	3.04
Interaction V x C	62.7	14	4.5	68.83	1.74
Interaction V x Y	17.4	14	1.2	19.14	1.74
Interaction C x N	129.3	4	32.3	496.93	2.41
Interaction V x C x Y	80.1	28	2.9	43.99	1.53
Within	0.5	3	0.2	2.58	2.65
Error	13.9	213	0.1		
Total	1121.1	287			

## Conclusion

The productivity of harvested grass biomass was found to be dependent on the cultivar used. Dry matter yield was also found to be strongly dependent on the weather conditions in each year of the trial, and particularly so during the periods of summer regrowth.

Significant differences in *Festulolium* and *Lolium x boucheanum* DM yield were found between the first, second and third years, with the greatest yield in the first harvest year.

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# The effect of root morphology on alfalfa yield in the spring regrowth period

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## Abstract

The goal of this study was to investigate the effect of the proportion of tap-roots to lateral roots on the yield of alfalfa. In an experiment with 15 alfalfa entries, the measurement and sampling were carried out in the spring regrowth period using 330 x 330 mm squares as the sampling units, with eight replicates of each alfalfa. The following parameters were measured: forage yield, number of plants, tap-root and lateral root weight and their weight proportions. The tap-root percentage ratio was positively related to yield and total weight of root. The decrease of stand density reduced significantly the tap-root ratio and total root weight. In the case of normalized total root weight per unit of area, the yield was not influenced by tap-root ratio or tap-root weight. It is possible to conclude that the proportion of tap-roots to lateral roots is not important for dry matter yield in cases of identical total root weight.

Keywords: alfalfa, tap-root, lateral root, yield

## Introduction

Generally, the morphology of alfalfa strongly influences its characteristics. Katić *et al.* (2003) reported that the morphological characteristics of alfalfa are significantly correlated with yield and dry matter quality. Lamb *et al.* (2000) showed that the productivity of alfalfa was influenced by root morphology. The effect of stand density on alfalfa root morphology was confirmed by Hakl *et al.* (2007). In that experiment Hakl used Jarka and several new candivars of alfalfa and showed that tap-root diameter was positively correlated with lateral root number and diameter and all of these variables were negatively correlated with the stand density. The effect of stand density was significant and explained about 15% of variability of measured parameters. It is possible to conclude that root morphology is important trait and is connected with stand density. We now report the results of an experiment at the Department of Forage Crops and Grassland Management which investigated effect of proportion of tap-roots to lateral roots on the stand productivity of alfalfa in the spring regrowth period.

## Materials and methods

A plot experiment with 15 alfalfa entries was established in the field of the Research Station of the Czech University of Agriculture in Červený Újezd in spring 2006. The mean annual temperature at this location is 7.7 °C, and the long-term annual sum of precipitation is 493 mm. The prevailing soil type is clay loam orthic luvisol with a neutral soil reaction.

The following 15 alfalfa entries were used: Czech varieties Pálava, Morava, Vlasta, Magda, Jarka, Zuzana, Jitka, Niva, Oslava, Kamila, the French variety Europe, and Czech candivars ŽE XLII, ŽE XLIII, ŽE XLVII, ŽE XLVIII. Sowing was by clear-seeding without herbicides, and the seeding rate for all entries was 7 million germination seeds ha<sup>-1</sup>. The experiment was a completely randomized design with eight replicates of each alfalfa entry and a plot size of 2.5 x 7.2 m. In spring 2007, the plants were sampled from an area 330 x 330 mm in each plot; the average depth of sampling was 200 mm. At the time of sampling, the height of stand was approximately 150 mm. The following parameters were measured for each sample: number of

plants, alfalfa dry matter yield, weight of tap-root (wTR) and lateral root (wLR). The total weight of root (TWR), tap-root (%TR) and lateral root percentage ratio (%LR) of weight were calculated. The relations among measured parameters were statistically evaluated by partial correlations where TWR was used as covariate. All statistical analysis were performed using Statistica 6.0 (StatSoft, Tulsa, OK, USA).

## Results and discussion

The stand density in alfalfa plots ranged widely from 11 to 366 plants m<sup>-2</sup> due to unfavourable weather in the establishment year. The main statistics for characteristics of all evaluated parameters are presented in Table 1. No significant differences in yield among alfalfa entries were detected so we did not include this factor in the following analyses.

Table 1. The main statistic characteristics of measured parameters (TWR = total root weight, wTR = weight of tap-root, wLR = weight of lateral root, %TR = weight percentage ratio of tap-root, %LR = weight percentage ratio of lateral root). SD = standard deviation, number of cases = 120.

	Mean	SD	Min	Max
Yield (g.m <sup>-2</sup> )	152.2	59.7	180.0	348.0
TWR (g.m <sup>-2</sup> )	121.4	44.4	30.0	235.6
wTR (g.m <sup>-2</sup> )	94.2	41.4	21.5	204.3
wLR (g.m <sup>-2</sup> )	27.1	13.7	36.3	66.3
%TR (%)	76.1	12.1	38.8	96.3
%LR (%)	23.9	12.1	3.7	61.2
Density (pcs.m <sup>-2</sup> )	92	55	11	366

Table 2 presents selected correlations between yields or stand density and measured parameters. In the case without covariates, tap-root weight and percentage ratio were significantly positively correlated with alfalfa yield and stand density but no significant relationship was detected between yield and weight of lateral root. The lateral root percentage ratio was negatively correlated with yield and stand density, which is in accordance with Hakl *et al.* (2007) about the negative effect of stand density on lateral root number and diameter.

Table 2. Correlation matrix and partial correlation matrix, including covariate among selected parameters (TWR = total root weight, wTR = weight of tap-root, wLR = weight of lateral root, %TR = percentage ratio of tap-root weight, %LR = percentage ratio of lateral root weight). Correlations significant at  $P < 0.05$  are in italics.

	Yield	TWR	Density	Yield	Density
Yield	1.0			1.0	
TWR	0.66	1.0		-	-
wTR	0.66	0.95	0.70	0.13	0.42
wLR	0.15	0.36	-0.07	-0.13	-0.42
%TR	0.34	0.36	0.48	0.14	0.35
%LR	-0.34	-0.36	-0.48	-0.14	-0.35
Density	0.59	0.63	1.0	0.30	1.0
Covariate		-		TWR	

The total weight of root (TWR) was positively related to yield. It corresponds with the findings of Lamb *et al.* (2000) on root morphology having an influence on alfalfa productivity. We excluded the effect of TWR by including it as a covariate and these normalized results can be seen in the right side of Table 2. It seems that the positive effect of tap-root parameters on yield was given only by higher weight of tap-root. In the case of normalized TWR, the

stands with higher density provided a higher yield. The relation between density and root morphology parameters was very similar as in the case without covariate.

## Conclusions

It is possible to conclude that alfalfa root distribution significantly influenced forage yield. The tap-root ratio positively related to yield and total weight of root. The decrease of stand density reduced significantly tap-root ratio as well as total root weight. In the case of normalized total root weight, the yield was not influenced by tap-root ratio or weight. It is possible to conclude that the proportion of tap-roots to lateral roots is not important for dry matter yield in cases of identical total root weight.

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# Importance of polyploidy for forage and seed productivity and forage quality of perennial ryegrass

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## Abstract

Two circular (radial) trials were carried out at the Institute of Forage Crops during the 2003-2005 period. Six populations (two Bulgarian diploids (D) and their two tetraploid (T) analogues of breeding populations, and the Belgian varieties Ritz (D) and Roy (T) as references) were involved in each trial under both irrigated or rainfed conditions in four replicates – two for forage and two for seed productivity measurements. This trial model is extremely economical, because in a small area it is possible to study a large number of individual plants and genotypes in a range of experimental environments, allowing the measurement of phenotypic (ecological and genotypic) dispersion. Polyploidy has a positive effect on forage and seed productivity and forage quality and is particularly suitable in perennial ryegrass breeding, where the main product is quantity and quality of above-ground biomass as animal feed.

## Introduction

The polyploidy status of plants has adaptive importance in species evolution. Polyploids arise in the periphery of gene centres and in that way the species broadens its area of distribution. Bulgaria is situated on the borders of two gene centres (Mediterranean and Caucasian), where biodiversity is at a maximum. Polyploidy is very valuable in cases where the vegetative plant organs are used as yield, so polyploids are very promising in forage crops. Perennial ryegrass is the most desirable forage species and is the key component of the most productive pastures. There are no naturally occurring polyploid types. In the 1960s, induced tetraploid varieties were first developed in Holland and later by other plant breeding centres. The percentage of tetraploids in variety lists for perennial ryegrass is increasing and that underlines their high importance for modern farm production. The high level of water soluble carbohydrate (WSC) is necessary in grasses to maintain persistency and also for cold and drought resistance. A significant positive correlation has been established between digestibility, intake and WSC (Halling *et al.*, 2004). Plant breeders pay attention to seed yield but it is difficult to combine high forage yield and persistence with high seed yield. The seed size of tetraploids is 40% larger (Najda, 2004) and their yield is 25% higher than diploids (Van Bockstaele, 1998). The aim of the study was to establish the effect of polyploidy on forage and seed productivity and on forage quality in a range of environments in order to test adaptation.

## Materials and methods

Two circular (radial) trials were carried out at the Institute of Forage Crops, Pleven, Bulgaria in the 2003-2005 period. Six populations (two Bulgarian diploids (D) and their two tetraploid (T) analogs of breeding populations and the Belgian varieties Ritz (D) and Roy (T) as references) were involved in each trial under both irrigated or rainfed conditions in four replicates – two for forage and two for seed productivity measurements. The tetraploids were obtained from DvP-Melle, Belgium within the framework of bilateral cooperation with IFC-Pleven, Bulgaria and the C<sub>3</sub> generation was used in the trials. Forty plants of each population were measured for forage productivity and forty for seed productivity in each trial. The individual plants were

established as seedlings in autumn 2003. Twenty plants per population were situated along the radius (20 cm distance within the row) of the circle. In the centre of the circle plant density was very high and competition very strong but at the edge of the circle there was no competition among the plants. All populations survived the first winter. 2004 was the first year for measurements of forage (4 cuts for BG and 3 for BE origins) and seed production. Next winter there was some winter damage. This was less for the Bulgarian populations than for the Belgian varieties and 2 cuts for forage production were carried out in 2005. Seed was harvested each year in duplicate. Rank analysis was carried out according to total average data over all the environments.

## Results and discussion

Dry matter productivity (g per plant) by environments and years for a total six cuts is given in Table 1. 2004 was nearly typical for our climatic conditions, but 2005 was exceptionally wet – just one irrigation was applied. Rainfed conditions had a detrimental effect on Belgian varieties in 2004 because of water deficiency, and they produced 3 cuts for forage in contrast to 4 cuts for the Bulgarian populations. In 2004, the Belgian varieties had higher forage productivity under irrigation, while the Bulgarian populations had better forage productivity in rainfed conditions. In 2005 the period from June to September was rather wet. During this third year of the established sward, forage productivity generally decreased in all populations, but the Bulgarian populations yielded better than reference varieties and the rainfed conditions were more favourable. Ranking of populations was carried out and all tetraploid variants were higher for forage productivity than their respective diploids.

Seed productivity (g per plant) by environments and years is given in Table 2. The highest seed productivity was obtained under irrigation, where SBG, Ritz and Roy were the best. Roy, as a tetraploid, produced more seed than Ritz. Within the Bulgarian populations there was no evidence for better seed productivity in the tetraploids because of a negative correlation between forage and seed productivity ( $r = -0.5184$ ), reported also by Razec *et al.* (2003).

Chemical composition (crude protein (CP), crude fibre (CF), water soluble carbohydrate content (WSC) and dry matter digestibility (DMD)) are presented in Table 3 as data averaged over all environments, years and cuts. Ranking was carried out and all tetraploid variants had more favourable forage quality characteristics than the respective diploids. The same conclusions were drawn by Fulkerson *et al.* (2004). Tetraploids possess a lot of advantages: darker green leaf colour, faster germination and sward establishment, greater 1000-seed weight, better leafiness, taller plants, higher forage and seed productivity, higher water and WSC content in the cells, and higher DMD and intake by animals, compared to diploids (Baert and Reheul, 1997; Van Bockstaele, 1998; Connolly, 2001 and Reheul *et al.*, 2003) and their development is a great achievement in perennial ryegrass breeding (Camlin, 1997).

Table 1. Dry matter productivity (g per plant) of diploid and tetraploid perennial ryegrass populations and varieties under irrigated and rainfed conditions (2004-2005).

Populations and varieties under irrigated and rainfed conditions (2004-2005):							
DM Productivity, g per plant							
Variant	Population, variety	Irrigated conditions		Rainfed conditions		Total	Rank
		2004	2005	2004	2005		
1	NBG	66.09	39.08	76.03	40.82	222.02	3
2	NBG - T	57.32	40.30	81.05	69.42	248.10	1
3	SBG	73.77	36.55	73.40	24.14	207.86	4
4	SBG - T	85.90	31.18	70.54	53.57	241.19	2
5	Ritz	84.74	23.80	58.25	27.14	193.93	6
6	Roy - T	87.09	27.55	61.16	28.13	203.94	5
Mean		75.81	33.08	70.07	40.54	219.50	
SD		12.24	6.64	8.79	17.93	21.57	



Table 2. Seed productivity (g per plant) of diploid and tetraploid perennial ryegrass populations and varieties under irrigated and rainfed conditions (2004-2005).

Seed productivity, g per plant							
Variant	Population, variety	Irrigated conditions		Rainfed conditions		Total	Rank
		2004	2005	2004	2005		
1	NBG	4.8	2.95	5.4	3.87	17.06	5
2	NBG - T	6.1	2.73	3.0	2.21	14.04	6
3	SBG	14.7	5.79	11.8	3.07	35.36	1
4	SBG - T	5.3	4.63	7.2	4.74	21.87	3
5	Ritz	9.5	4.77	4.5	2.7	21.47	4
6	Roy - T	9.6	3.25	5.7	5.06	23.61	2
Mean		8.3	4.0	6.3	3.6	22.2	
SD		3.75	1.22	3.04	1.14	7.33	

Table 3. Chemical composition and dry mater digestibility (g kg<sup>-1</sup> DM) of diploid and tetraploid perennial ryegrass populations and varieties (average values from all conditions and cuts for (2004-2005) period).

Variant	Population, variety	CP	CF	WSC	DMD	Rank
		g kg <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	g kg <sup>-1</sup>	
		DM	DM	DM	DM	
1	NBG	172.6	208.9	63.0	616.2	5
2	NBG - T	189.1	199.7	66.0	636.1	2
3	SBG	161.5	209.8	62.3	593.5	6
4	SBG - T	162.6	206.5	67.0	616.3	4
5	Ritz	187.4	227.3	31.4	624.5	3
6	Roy - T	185.0	211.1	44.2	652.5	1
Mean		176.37	210.55	55.65	623.18	
SD		12.52	9.15	14.51	20.03	

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# Genotype x environment influence on first-cut dry matter yield stability of annual ryegrass cultivars

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## Abstract

Dry matter yield stability of 8 annual ryegrass (*Lolium multiflorum* var. *westerwoldicum* Lam.) varieties at the first cut was studied in three different locations of Lithuania (Dotnuva, Joniškėlis, and Perloja) during 2006-2007. Sites were on a light loam Endocalcari-Epihypogleyic Cambisol (Dotnuva), an Endocalcari-Epihypogleyic Cambisol (Joniškėlis) and an Hapli-Albic Arenosol (Perloja). The weather conditions differed in experimental years: 2006 was droughty; in 2007 there was enough moisture.

A good variety must not only give a high dry matter (DM) yield but also it should be stable, i.e. the yield must be high in different growing environments. Genotype had significant ( $P < 0.05$ ) effect on DM yield under all weather conditions in different locations. The varieties 'Rapid' and 'Elunaria' produced the highest DM yield at the first cut in both years at all three locations (1.56 and 1.43 Mg ha<sup>-1</sup> in 2006; 1.56 and 1.15 Mg ha<sup>-1</sup> in 2007) and there was low variance of stability (0.004 and 0.032) compared with the standard Lithuanian variety 'Varpė'. The variety 'Pollanum' produced the lowest DM yield in both years (0.92 Mg ha<sup>-1</sup> in 2006, and 0.85 Mg ha<sup>-1</sup> in 2007). The varieties 'Rapid' and 'Elunaria' gave the highest DM yield at the first cut under different environmental conditions.

Keywords: annual ryegrass, dry matter yield, stability

## Introduction

As agriculture is becoming more and more intensive, production of high quality feed, even under unfavourable growing conditions and without any damage to surrounding ecosystems, is crucial. Ryegrass is one of the most valuable forage grasses. The genus ryegrass includes ten species, but only three, perennial (*Lolium perenne* L.), Italian (*L. multiflorum* Lam.), and annual (*L. multiflorum* var. *westerwoldicum* L.) are used in agriculture. Annual ryegrass is a high quality cattle feed that is available through all summer until late autumn. It can be grown as a pure crop and in mixtures for green forage. Annual ryegrass can also be used for sowing in places where perennial grasses or winter crops have been killed by frost. Annual ryegrass grows rapidly, and under favourable growing conditions up to 3 yields can be obtained. The first cut gives the best yield - up to 50% of the total annual yield (Lazauskas, 1998; Truzina, 2003).

A very important property of forage grasses is their ability to give stable high dry matter yields under different environmental conditions (Bidinger *et al.*, 1996). The level of productivity depends on the genetic potential of the variety, which is controlled by the response limits of the genotype. Yield stability depends on the ability of a variety to respond to environmental conditions. There are varieties with high adaptability, which give stable but lower yields under different environmental conditions, whereas varieties with a low adaptability, yield high in favourable and low under unfavourable conditions (Borojevic, 1990).

The aim of our study was to investigate annual ryegrass first-cut yield stability according to a genotype x environment model.

## Materials and methods

Eight varieties of annual ryegrass (*Lolium multiflorum* var. *westerwoldicum* Lam.) were used: 'Varpe', 'Rapid', 'Druva', 'Lifloria', 'Pollanum', 'Elunaria', 'Imperio' and 'Lirasand'. The experiments were set up in three different sites of Lithuania (Dotnuva, Joniškėlis, Perloja) during 2006-2007. Dotnuva (55°23'N, 23°57'E) was on a light loam Endocalcari-Epihypogleyic Cambisol; Joniškėlis (56°07'N, 24°10'E) was on a Endocalcari-Epihypogleyic Cambisol; and Perloja (54°13'N, 24°25'E) was on a Hapli-Albic Arenosol. In each location, the 8 varieties were planted in 6.25 m<sup>2</sup> test plots using a randomized complete block design with 4 replications, to a genotype x environment model (Eberhart and Russel). Sowing was made on May 2-5. Cutting was performed when annual ryegrass was at the heading stage.

The weather conditions in all experimental sites were similar, but there were differences between years, especially in May, which was droughty in 2006 and sufficiently wet in 2007. As no significant differences were found between the locations, mean values of weather conditions for the three sites are presented (Figure 1).

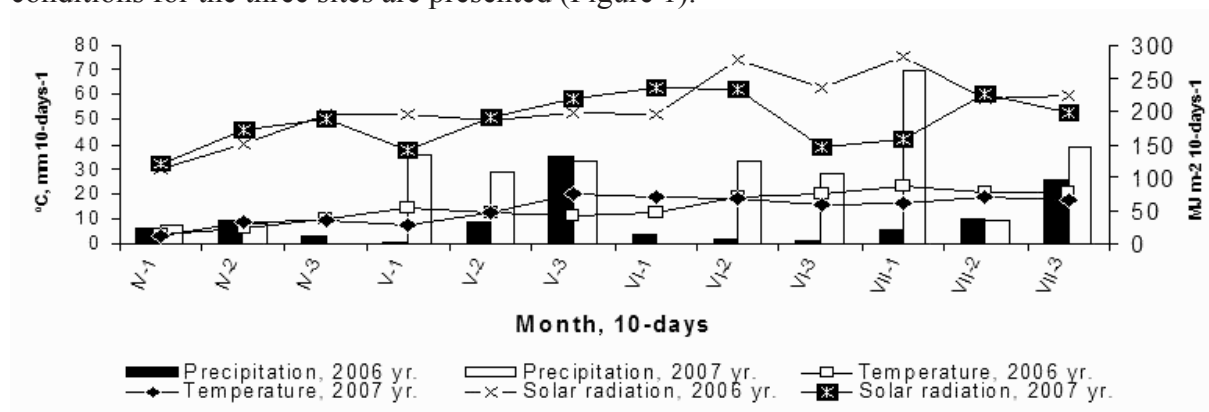


Figure 1. Weather conditions during 2006-2007.

The stability analysis computer software YIELDSTAB developed by P. Tarakanovas in the Visual Basic of Application as macro program to run in EXCEL was used. This program includes regression (Brewbaker, 1995) and stability analysis (Kang, Magari, 1995).

## Results and discussion

Annual ryegrass requires moisture and warmth (Lazauskas, 1998). Investigations show that DM yield in 2006 was insignificantly ( $LSD_{05} = 0.19$ ) higher than in 2007, although there was less precipitation during May and June (except for May third 10-days), but the air temperature and solar radiation was higher in May first 10-days and June second-third 10-days (Figure 1). These factors could have determined the timing of the first cut: in 2006 it was about 2 weeks earlier than in 2007 (08.07-11.07 and 23.06-28.06).

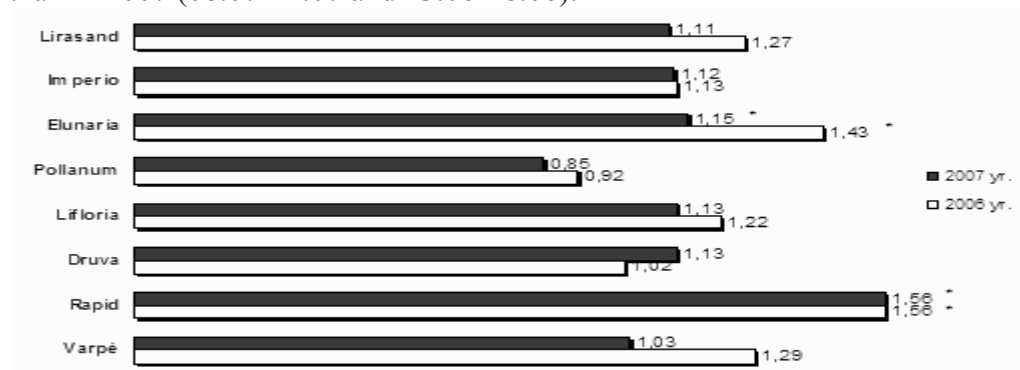


Figure 2. Annual ryegrass dry matter yield (Mg ha<sup>-1</sup>) at the first cut in 2006, 2007. (\* $P < 0.05$ )

Investigations of environmental influence showed that DM yield at the first cut was significantly higher ( $LSD_{05} = 0.56$ ) at Joniškėlis than Dotnuva and Perloja, in both years. Annual ryegrass DM yield is determined not only by the weather or environmental conditions, but also by a genotype. Genotype had a significant ( $P < 0.05$ ) effect on DM yield under all weather conditions at the different locations. The varieties 'Rapid' and 'Elunaria' produced the highest DM yield ( $1.56 \text{ Mg ha}^{-1}$  and  $1.43 \text{ Mg ha}^{-1}$ ) in all three locations, compared to the standard variety 'Varpė'. The variety 'Pollanum' gave significantly ( $P < 0.05$ ) low dry matter yield ( $0.92 \text{ Mg ha}^{-1}$ ) (Figure 2).

In 2007 all varieties investigated produced higher DM yield than the standard variety 'Varpė', except for the variety 'Pollanum' ( $0.85 \text{ Mg ha}^{-1}$ ). 'Rapid' and 'Elunaria' had significantly ( $P < 0.05$ ) higher DM yield,  $1.56 \text{ Mg ha}^{-1}$  and  $1.15 \text{ Mg ha}^{-1}$  respectively (Figure 2).

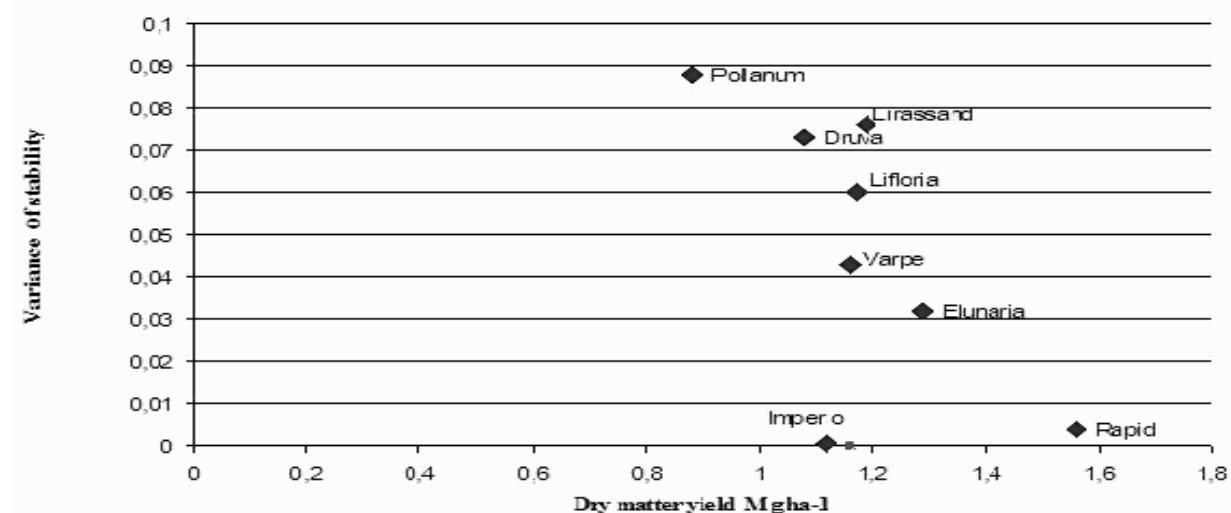


Figure 3 Scatter diagram for dry matter yield at the first cut of annual ryegrass and variance of stability 2006-2007.

A good variety must not only give a high DM yield but also to be stable, i.e. the yield must be high in different growing environments. This research showed low stability variance in the different locations of Lithuania for all varieties investigated. 'Rapid' and 'Elunaria' had low stability variance (0.004 and 0.032) and significantly higher DM yield in 2006-2007. 'Pollanum' had the highest stability variance (0.088), and the lowest DM yield (Figure 3).

## Conclusions

Genotype had a significant ( $P < 0.05$ ) influence on DM yield under all weather conditions in the different locations. The varieties 'Rapid' and 'Elunaria' gave the highest stable DM yield at the first cut under different environmental conditions. The variety 'Pollanum' produced the lowest, but most stable, DM yield of all varieties tested.

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# The influence of management and exploitation of grasslands on the differentiation of their typological structure, biodiversity and productivity

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## Abstract

Between the years 2000–2006 experiments were carried out to study the influences of different ways of managing and exploiting permanent grasslands (mowing – grazing; harvest frequency; fertilization) on the differentiation of their typological structure, biodiversity and productivity in the Bohemian Forest (650 m a.s.l.). Three years after the beginning of the experiments, the original single stand type with *Dactylis glomerata* as the dominant species had changed and grown into three varied stand types, and after six years into five varied stand types. Homogeneous groups of more productive stands (at  $P = 0.05$ ) with lower than mean yield variability influenced by the particular year ( $V_y \leq 17.75\%$ ) included (1) stands cut twice, both with and without fertilization, and (2) stands grazed three times, with significantly positive values of species richness (twice-cut growths) or diversity (swards grazed three times).

Keywords: grassland management, typological structure, biodiversity, yield

## Introduction

The system and intensity of treatment and exploitation significantly influences the richness and diversity of species, total stand composition and productivity of grasslands (Hofmann *et al.*, 2001). Considering the influence on the characteristics of grassland dynamics (Klimeš, 2000; Klimeš and Voženílková, 2001), it is necessary to solve the question of the harmonization of grassland production and the non-production functions, while applying combined management systems and utilization.

## Materials and methods

Between the years 2000–2006 experimental studies were carried out on the influences of different types of management and exploitation (Table 1) of permanent grasslands on the differentiation of their typological structure, species richness, diversity and productivity. The experiments were conducted 4 times and the study area was in the sub-mountain region of the Bohemian Forest (650 m a.s.l., mesophyte locality). Phytocenological analyses were made from relèves, recorded each year before the first harvest. Species richness (S) is expressed as total number of species of vascular plants. Species diversity was evaluated according to the Simpson Index (D). Experimental data were evaluated by methods of variation analyses and analysis of variance.

## Results and discussion

In the course of the experimental period the original stand type, with *Dactylis glomerata* as the dominant species, changed and after three years had evolved into three different stand types, and after six years into five types (Table 1). Based on complex evaluation (S, D,  $HG_{0.05}$ ), the most suitable stands are particularly those cut 3-times, both with and without fertilization, followed by the stands cut twice without fertilization. The least suitable regime, with regard to both values (S, D), seems to be the utilization of fertilized stands only in two pasture cycles (Table 1, 2).

Table 1. The survey of experimental variants, changes of typological structure of dominant species group (dominant species marked in bold) and of differentiation of species diversity according to the Simpson Index (D).

Method of Utilization (i)	f (ii)	Fertilization (iii)	Treatment Frequency (i)(ii)(iii)	Year			$\bar{D}$	HG <sub>0.05</sub> (D)
				2000	2003	2006		
Mowing (M)	2	-	M2/0	<i>Dag-Fep-Trf</i>	<i>Trr-Php-Fep</i>	<i>Trf-Trp-Taxr</i>	12.31	*
		NPK	M2/NPK	<i>Dag-Fep-Trf</i>	<i>Php-Fep-Pop</i>	<i>Trf-Php-Dag</i>	9.89	*
	3	-	M3/0	<i>Dag-Fep-Trf</i>	<i>Trr-</i>	<i>Trp-Taxr</i>	13.56	*
		NPK	M3/NPK	<i>Dag-Fep-Trf</i>	<i>Trr-Pop-Php</i>	<i>Taxr-Trf</i>	13.21	*
Grazing (G)	2	-	G2/0	<i>Dag-Taxr</i>	<i>Trr-Dag-Pop</i>	<i>Taxr-Pll</i>	12.28	*
		NPK	G2/NPK	<i>Dag-Taxr</i>	<i>Dag-Pop-Php</i>	<i>Php-Pop-Dag</i>	10.63	*
	3	-	G3/0	<i>Dag-Taxr</i>	<i>Trr-Agc</i>	<i>Trr-Pop-Agc</i>	13.37	*
		NPK	G3/NPK	<i>Dag-Taxr</i>	<i>Dag-Trr-Pop</i>	<i>Taxr-Pop-Fep</i>	13.32	*

f – frequency of utilization; NPK – application of 100 kg N ha<sup>-1</sup> +PK; *Dag* – *Dactylis glomerata*, *Fep* – *Festuca pratensis*, *Trf* – *Trisetum flavescens*, *Taxr* – *Taraxacum sp. ruderalia*, *Php* – *Phleum pratense*, *Pop* – *Poa pratensis*, *Trr* – *Trifolium repens*, *Agc* – *Agrostis capillaris*, *Pll* – *Plantago lanceolata*; HG<sub>0.05</sub> – homogeneous groups ( $P_{0.05}$ ).

Table 2. Species richness (S) and yield ( $Y = t \text{ DM ha}^{-1}$ ) on experimental stands.

Treatment	$\bar{S}$	HG <sub>0.05</sub> (S)			V <sub>s</sub> (%)	$\bar{Y}$	HG <sub>0.05</sub> (Y)		V <sub>y</sub> (%)
M2/0	31.57	*	*	*	9.31	6.38	*	*	7.52
M2/NPK	28.43	*	*	*	8.09	7.91	*	*	16.60
M3/0	33.29	*	*	*	15.71	4.80	*	*	20.06
M3/NPK	29.71	*	*	*	12.99	6.56	*	*	23.84
G2/0	27.71	*	*	*	6.82	6.07	*	*	20.21
G2/NPK	26.57	*	*	*	10.61	7.16	*	*	20.82
G3/0	25.43	*	*	*	9.87	5.11	*	*	16.58
G3/NPK	26.86	*	*	*	11.24	6.18	*	*	16.38

V<sub>s</sub> (%), V<sub>y</sub> (%) – variation coefficients; HG<sub>0.05</sub> - homogeneous groups ( $P_{0.05}$ ) for S and Y.

Table 3. Yield ( $Y = t \text{ DM} \cdot \text{ha}^{-1}$ ) and its variability (V<sub>y</sub> %) for one-sided and combined exploitation (½ area mowing [M] + ½ area grazing [G]) with marked effect of combined exploitation on the change of total variability of yields [ $\Delta V_y$  (%)].

Treatment combination	Y	HG <sub>0.05</sub>	V <sub>y</sub> (%) at comb. M + G		HG <sub>0.05</sub>	V <sub>y</sub> (%) for one-sided exploitation		$\Delta V$ (%) for combined exploitation compared with one-sided exploitation	
			M + G	HG <sub>0.05</sub>		M	G	M	G
M3 + G3	5.66	*	18.36	*	*	21.96	16.48	-3.60	+1.88
M3 + G2	6.15	*	18.21	*	*	21.96	20.52	-3.75	-2.31
M <sub>1</sub> G*	6.27	*	(17.76)	*	*	17.02	18.50	-	-
M2 + G2	6.88	*	12.12	*	*	12.07	20.52	+0.05	-8.40
M2 + G3	6.40	*	8.43	*	*	12.07	16.48	-3.64	-8.05
M2/0 + G3/0	5.75	*	6.57	*	*	7.52	16.58	-0.96	-10.02
M2/NPK + G3/0	6.51	*	6.61	*	*	16.60	16.58	-10.01	-9.98
M2/0 + G3/NPK	6.28	*	7.50	*	*	7.52	16.38	-0.03	-8.87
M2/NPK + G3/NPK	7.05	*	13.06	*	*	16.60	16.38	-3.56	-3.31

\*M<sub>1</sub>G - exceptionally not combination (M + G), but set of all variants; HG<sub>0.05</sub> - homogeneous groups ( $P_{0.05}$ ).

Homogenous groups of more productive stands (at the level  $P = 0.05$ ) with lower than mean yield variability influenced by the particular year ( $V_y \leq 17.75\%$ ) include (1) stands cut twice, both fertilized and without fertilization, and (2) stands grazed three times, and there were



significantly positive values for species richness (on the twice-cut stands) or diversity (on the three times grazed swards).

On the basis of the study of methodological questions concerning the effects of particular years on species richness (S) variability, and on grassland yield (Y) variability (Klimeš, 2000; Klimeš and Voženílková, 2001) the verified variants displayed tendencies towards prevailing sinusoid oscillations in terms of both species richness and yields. In cut and grazed stands, adverse tendencies in yield oscillations were recorded, which was shown by yield stabilization in particular years on condition that a part of the area with synecologically similar stands is exploited by means of mowing or grazing. This stabilization of yields was especially significant in twice-cut stands with three cycles of grazing (Table 3).

## Conclusions

In considering the harmonization of grassland production and the non-production functions of grasslands and related problems, it is necessary to carry out long-term experiments which include a broad range of variants under different treatment and exploitation regimes. From this perspective, when considering synecologically similar stands, it is appropriate to apply a combined exploitation consisting of cutting twice a year (on half of the area) and pasture with three grazing cycles per year (on a half of the area).

## Acknowledgement

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# The problems and perspectives of the grassland in Belarus

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## Abstract

In Belarus, grassland occupies more than one-third of the agricultural land. It provides the livestock with forage during the pasture period (150-160 days per year) as well as during winter (November to May). Socio-economic aspects and the location of the country, as well as the consequences of the regional and global ecological problems (melioration of the swamps, the explosion at the Chernobyl nuclear power station) have greatly affected the conditions for the development and utilization of grassland. The grassland on the meliorated peat soils, and the preservation of the natural swamps and forests, all have enormous ecological impacts on sustaining biospheric processes. Taking into account all the factors, it is crucial to work out and adapt the strategies for the development and utilization of the grassland, paying attention to the soil and climate conditions and ecological peculiarities of the country.

Keywords: environment, grassland, productivity, soil

## Discussion

Belarus is a small state with a population of 10 million people, 28% of whom live in the rural areas. The historical past, as well as regional and global ecological problems, have affected the socio-economic development of the country. As a result of the explosion at Chernobyl nuclear power station in 1986, one-fifth of the territory of the country was subject to radioactive contamination, namely 14.9% of the agricultural land and 21.7% of the forestry. The main purpose of the agricultural management on the contaminated land is to bring the agricultural products containing radionuclides within the accepted ecological standards.

Belarussian scientists conducted research to find out the range of the crops and species characterized by the minimum capacity to accumulate radionuclides, the methods to cultivate land, the types of fertilization and the typical schemes of rotation according to the level and character of the radioactive contamination of the soils. The protective measures conducted just after the Chernobyl catastrophe made it possible to reduce the accumulation of radionuclides in the crops by 8 to 10 times (Shevchuk and Gurachevskiy, 2006).

It is necessary to implement measures to regulate the water regime and regenerate the pastures and meadows (used by individual farmers to provide grazing), which became excessively wet and lost their productivity. The main problem is that the meliorated soils revert into swamps. If that is the case, the radioactive contamination of the forage increases, consequently polluting the milk and meat. As a result, the maintenance of the melioration system in the areas whose soil is characterized by radioactive contamination is of strategic importance.

In Belarus 65.8% of the territory was characterized by excessive soil wetness, and the existence of the swamps has prevented intensive development of the country for many years, as farms would lose up to 40% of the harvest of the agricultural crops owing to the unregulated water regime of the land. In the period from 1960 to the 1980s, a wide-ranging programme of melioration was conducted. It made it possible to increase the productivity of the meliorated area by 5-6 times. The large areas of meliorated land made it possible to increase the production of agricultural products by at least up to 30% every year and build a relatively developed social infrastructure (Gusakov, 2001). By now, 2,900 ha of the area has



been meliorated, and 40% of the meliorated soils are utilized as arable land, with 60% as meadow. However, owing to the long and inappropriate utilization of the meliorated peat soils for arable crops and cereals, more than 300,000 ha was first anthropogenically transformed into organico-mineral soil. This requires us to investigate the transformed peat soils and work out special methods for their utilization (Lishtvan and Yaroshevich, 2001).

The after-effects of the wide-ranging melioration and inappropriate utilization of the meliorated land has resulted in the changing of the climatic and ecological conditions. Moreover, biochemical cycles of carbon, nitrogen and other biogenic elements have changed. As a result, water, air and agricultural products have become contaminated by the elements that were released after the decomposition of the peat. Eutrophication of the natural water reservoirs has increased. Despite the wide-ranging melioration and the intensive utilization of the peat soils, especially in the south of the country, a considerable amount of the naturally-sustained swamps (1,706 ha) has been preserved, and overflow lands have also been turned into swamps, which are of great importance for European and global biospheric processes.

The reduction of the use of peat fertilization and the decrease of the livestock numbers led to reduced applications of organic fertilizers. As a result, the fertility of the soils has dropped. Now the fertility of the arable land is maintained at a medium average level (humus 2.25%, P 178 mg kg<sup>-1</sup>, K 190 mg kg<sup>-1</sup>, pH 5.98), whereas the fertility level of the grassland is low. There is evidence of reduced phosphorous fertilization (we have to import phosphorous) and over the last 10 years there has been the tendency of decreased reserves of phosphorous in the soil. The resources of Belarus do not make it possible to develop expensive agriculture. The deficiency of the budget requires us to utilize a range of resource-saving and cost-effective technologies designed to maximize local natural conditions for the most effective use of their potential.

The scientists and meteorologists worked out a new adaptive strategy to develop the national agriculture, and the grassland in particular. According to the conducted soil and ecological zonation, there are three provinces of the country (north, central and south) and five regions of agricultural specialization. The basic food, feed and industrial crops are cultivated throughout all the regions. The scientists advise that we should increase the cultivation of the drought-resistant plants (potatoes, corn, sorghum and others) to reduce the negative influence of the frequent droughts, whereas spring crops and perennial grasses should occupy the main part of the cultivated crop areas on the soils with high soil moisture levels (Privalov *et al.*, 2007).

Perennial grasses occupy more than 20% of the arable land. The main cost-effective reserve to increase the production of forage and protein is to alter the species composition of the grasses and change over the annual use of clovers and biannual use of the clover-grass mixture. The long utilization of the meadows without renovation (which requires a lot of investment) led to the decrease of their productivity. Under these conditions, they developed the technology of sod sowing into the aged swards to reduce the inputs by up to 30%.

Now there are more than 1,400 ha of these pastures in Belarus, including 1,100 ha of cultivated pastures. In the last 10 years the areas of pasture renovation have reduced by more than two-fold, and their productivity has decreased. As a result, the area of poor pastures has increased. It is preferred to use legume species in the sward, or in sod sowing, to increase the productivity of the pastures as a resource-saving technology for grassland.

Every year yields of 6,500-7,800 kg ha<sup>-1</sup> of cereals are obtained in the country. Cereals amount more than 50% of the sown area. Nationally selected varieties of cereals are used to sow the majority of the land (more than 70%). Now there is a 30% overspend of concentrate feeding because of imbalance of protein. The enlargement of the sown areas and increase of the yield of grain legumes can help to resolve the problem. In 2006 the areas sown to peas and vetch occupied more than 60% of the whole sown areas of grain legumes. Taking the long

view, the sown areas for these crops will be increased. The use of tall-growing plants and slow change in the use of old varieties for new ones are the main problems concerning crops. In 2005-2006 the yield of green corn averaged 20,000 kg ha<sup>-1</sup>, of which grain comprised an average of 4,000 kg ha<sup>-1</sup>. That means that the potential yield of corn is not fully realized. Virtually, the whole volume of the silage is made from corn, and the areas for grain are being enlarged. According to the scientists and practitioners it is necessary to upgrade the intensification of the technologies, given the need to save energy by locating the corn fields reasonably near stock-raising corporations and farms.

Seed breeding has been carried out to produce twelve species of perennial grasses and eight species of legumes. A comparative study was conducted on peat and mineral soils in Karelia of various species and varieties of perennial grasses and legumes, which were selected in Belarus and abroad, and confirmed high genetic potential of the national varieties (Kulakovskaya, 1997). The main problems in seed-breeding of the perennial grasses are to preserve the undercover sowings during the winter.

## Conclusion

The after-effects of the melioration and radioactive contamination following the explosion at the Chernobyl nuclear power station, and the decrease of livestock numbers, have generated serious problems in utilization and management of the grassland in Belarus. Measures have been taken to solve the existing problems, in terms of the perfection of the specialization of the agriculture, as well as the change of the structure of agricultural land given the characteristics of the soil, climate and ecological conditions in Belarus. There has been an increase in livestock numbers, and increased productivity of the meadows and pastures has been achieved due to the use of resource-saving but effective technologies. Unproductive land has been excluded from the agricultural rotations to create bio-energy plantations and afforestation, and conservation of the meliorated swamps which had ceased achieving their natural biospheric functions.

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## Yields of the sward depending on utilization method and species tested in mixtures

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### Abstract

The aim of this work was to estimate the yielding of mixtures with *Festulolium loliaceum* and other species, depending on the utilization method. Permanent yielding during the vegetative season is the most important feature of grasslands. Breeders seek new genotypes characterized by high yield and adaptation to conditions of environmental stress. In the hybrid *Festulolium loliaceum* (*Festuca pratensis* x *Lolium perenne*), the basic aim of crossings between *Lolium* and *Festuca* is combination of the persistence, winter hardiness and drought tolerance of fescues with high yield and quality of ryegrasses. Investigations were carried out in eastern Poland in 2002-2006, years that were characterized by conditions of environmental stress. Two research factors were considered: (i) method of utilization (pasture – Limousin cattle grazing and simulated utilization – proportional number of cutting) and (ii) grass-clover mixtures with tested species (*Poa pratensis*, *Festulolium braunii*, *Festulolium loliaceum* – 2 strains, *L. perenne* and *F. pratensis*). The simulated sward had the highest yield in the first period of the studies, while the pasture sward had the highest yield in the years 2005-2006. Sward yield depended less on the species in the mixture. The studies confirmed the utility of *Festulolium loliaceum* strains to pasture mixtures in post-boggy habitats.

Keywords: *Festulolium loliaceum*, pasture and simulated utilization, yielding

### Introduction

Productivity of grassland depends on many factors including sward botanical composition, level of mineral fertilization, soil and climatic conditions (Grzegorzczak, 1989; Mikołajczak and Warda, 1997; Baryła, 2001; Baryła and Kulik, 2005). Grass communities react strongly to rainfall deficiency in a growing season. A most important requirement of grasslands, especially pastures, is the maintenance of stable yielding during the growing season. Many pasture studies involve simulated utilization (frequent cutting at pasture maturity), without other zoogenic factors. Moreover, plant breeders seek new genotypes characterized by higher yields and good adaptation to environmental stress, such as the hybrids of *Festuca pratensis* x *Lolium perenne* [*Festulolium loliaceum* (Huds.) P.V. Fourn] obtained at the Institute of Plant Genetics PAS in Poznań (Kulik *et al.*, 2004). The aim of this work was to estimate the yielding of mixtures with *Festulolium loliaceum* and the other species depending on the utilization method.

### Materials and methods

The studies were carried out in 2002-2006 in Didactic-Research Station in Sosnowica. A permanent grassland complex is located in the region of Wieprz-Krzna Channel (eastern Poland). Experiments were set up on a pasture area on organic soil (peat-muck soil) in 4-replication split-plot design. The first factor was the utilization method (pasture vs. simulated). Sward treatments comprised six sown grass-legume mixtures each with a 30% share of the tested species (1. *Poa pratensis* cv. SKIZ, 2. *Festulolium braunii* cv. Felopa, 3. *Festulolium loliaceum* I strain – spreading type, 4. *Festulolium loliaceum* II strain – erect type, 5. *Lolium perenne* cv. Solen and 6. *Festuca pratensis* cv. Skra). Constant components of

the sown mixtures were the following: *Phleum pratense* cv. Odra (35%), *Dactylis glomerata* cv. Areda (10%) and *Trifolium repens* cv. Romena (25%). The initial hybrids between tetraploid forms of *Festuca pratensis* and *Lolium perenne* were obtained at the Institute of Plant Genetics PAS in Poznań. Breeding materials were developed at the Szelejewo Plant Breeding station. During each of five years of studies fertilization was applied at rates of 75 kg N, 31 kg P and 75 kg K ha<sup>-1</sup>). The pasture swards were grazed by Limousin cattle, 5-6 times during each grazing season, while the simulated swards were cut at the same time and stage of pasture maturity. The area of individual pasture plots was 30m<sup>2</sup> and of the simulated plots 15m<sup>2</sup>. Before every regrowth an estimate of sward yield was made by mowing a 5.5m<sup>2</sup> area of each plot. Results of the dry matter yield measures were analysed using the Tukey test.

## Results and discussion

Dry matter yield of the pasture sward was different in different years of the studies and it depended on the method of utilization as well as on species composition of the sown mixtures. Significant differences between grazing and simulated utilization were noted in the years 2003-2006. The mowing sward was characterized by significantly highest yield at the 2nd regrowth in 2003 (simulated (s) – 1.83 Mg ha<sup>-1</sup>, pasture (p) – 1.01 Mg ha<sup>-1</sup>) and in 2004 (s – 1.73 Mg ha<sup>-1</sup>, p – 1.35 Mg ha<sup>-1</sup>) as well as in annual yield in 2004 (s – 10.02 Mg ha<sup>-1</sup>, p – 9.29 Mg ha<sup>-1</sup>). However, the pasture sward was characterized by being significantly the highest yielding at the 1st regrowth (p – 1.52 Mg ha<sup>-1</sup>, s – 1.23 Mg ha<sup>-1</sup>) and the 3rd regrowth (p – 1.57 Mg ha<sup>-1</sup>, s – 1.21 Mg ha<sup>-1</sup>) in 2003, at the 1st (p – 1.34 Mg ha<sup>-1</sup>, s – 0.85 Mg ha<sup>-1</sup>) and the 5th (p – 1.26 Mg ha<sup>-1</sup>, s – 1.09 Mg ha<sup>-1</sup>) regrowth in 2005, as well as at the 5th regrowth in 2006 (p – 1.11 Mg ha<sup>-1</sup>, s – 0.73 Mg ha<sup>-1</sup>) (Figure 1). The results show that the yielding of simulated and pasture sward was considerably differentiated in particular years of the studies.

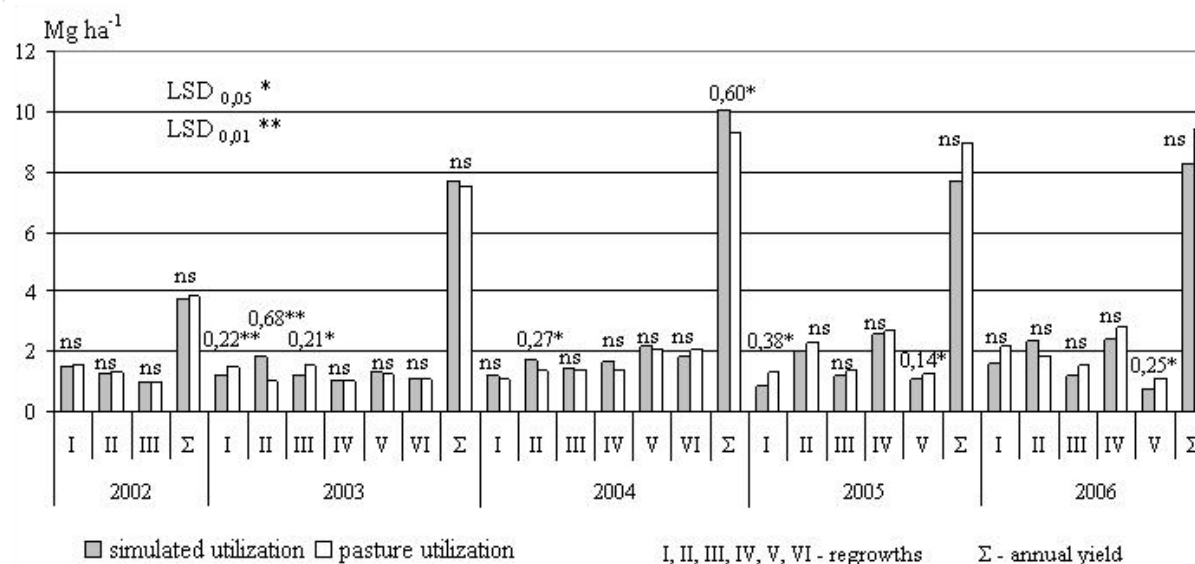


Figure 1. Average yields of the sward depending on the utilization method.

In the first period of the investigations it was noted that there was higher yielding in the simulated sward utilization, while the pasture sward had the highest yield in the years 2005-2006. Dry matter yield of the pasture sward was affected by random deposition of excreta by grazing cattle. The higher sward yields were connected with higher concentrations of macroelements in soil in the later years of the studies (Rogalski *et al.*, 2000). Sward yields depended slightly on the species tested in the mixture. The mixture of *Poa pratensis* and *Festuca*



*pratensis* was characterized by significantly the highest DM yield at the 1st regrowth and in the annual yield of 2003. In the 5th regrowth of 2003 it was noted that the significantly highest yielding mixture was with *Festulolium loliaceum* II, and in the 5th regrowth of 2006 it was the mixture with *Lolium perenne* (Figure 2). Yields of the mixtures with *Festulolium loliaceum* strains were comparable to the other tested mixtures and to results from other research (Grzegorzczuk, 1989; Mikołajczak and Warda, 1997; Baryła and Kulik, 2005). These studies carried out at the Didactic-Research Station in Sosnowica confirmed the utility of *Festulolium loliaceum* strains for pasture mixtures on peat-muck soil.

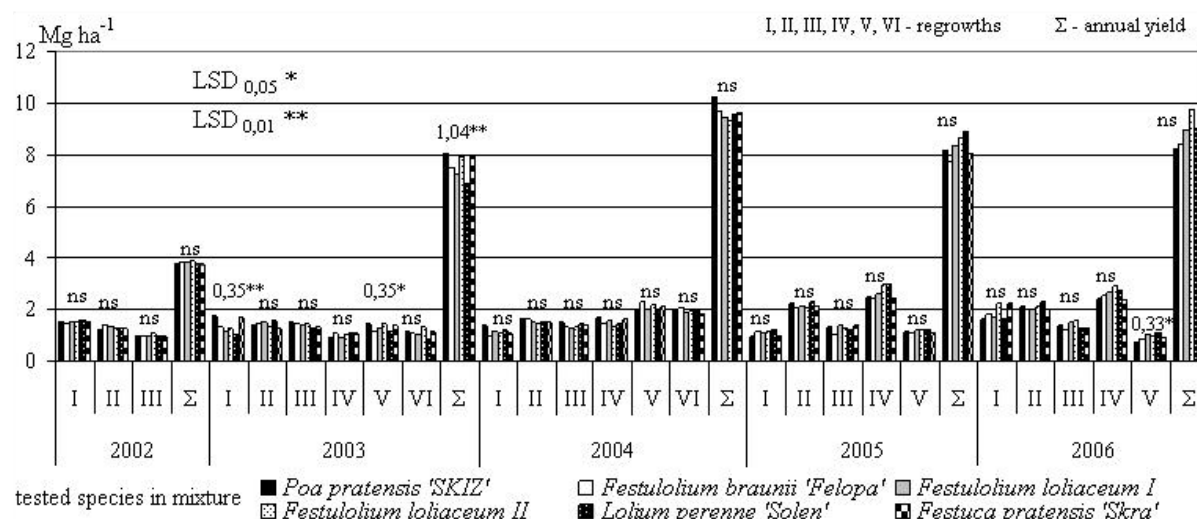


Figure 2. Average yields of the sward depending on species tested in mixture.

## Conclusions

- The simulated sward was significantly the highest yielding sward in the first period of the studies, while the pasture sward had the highest yield in the years 2005-2006.
- The studies confirmed the utility of *Festulolium loliaceum* strains for use in pasture mixtures in post-boggy habitats of eastern Poland.
- Sward yielding depended only slightly between the species tested in the mixtures.

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# Grass and legume productivity oscillations in a binary mixture

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## Abstract

The interaction between species in a grass-legume association provides the basis for large oscillations in their productivity. An experiment was carried out with the objective to investigate the effect of cutting regime on the year-by-year and seasonal variation in yield and botanical composition of a tall fescue-alfalfa mixture. The experiment was conducted in Drama, northern Greece. Plants of alfalfa (*Medicago sativa* v. ylikí) and tall fescue (*Festuca arundinacea* cv. Festorina) were cultivated in a 1:1 mixture, with four replications. Three cutting regimes were applied: a) one cutting at the stage of full maturity (namely uncut, C0), b) cutting at 7 cm above the soil surface (light cut, C7), and c) cutting at 3 cm above the soil surface (heavy cut, C3). Over a period of four successive years the above-ground dry biomass (AGDB) of each plot was separated into alfalfa and tall fescue and weighed. The results showed that increased cutting intensity decreases the annual and the total AGDB of the mixed swards. The botanical composition of the mixtures oscillated within and between growing seasons. The cutting improved the competitive ability of tall fescue, which successfully competed with alfalfa in this Mediterranean climate with irrigation.

Keywords: grass, legume, mixture, competition, productivity, cutting

## Introduction

Mixtures adapt better to a wide range of environments and utilize the resources better than monocultures, while mixture performance depends on the particular species interaction and environmental factors. The maintenance of species balance in a mixture is essential for sustainable mixtures. The balance is affected by the management practices, the more common of which is the cutting regime. The main effect of cutting is that it suppresses competitive dominance (Lazaridou and Vrahnakis, 2005).

Our objective was to investigate how the year-by-year and seasonal oscillations of the yield and botanical composition of a mixture of tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*) is affected by cutting regime.

## Materials and methods

A field experiment was conducted at the Tobacco Institute of Drama, northern Greece (41°09'N., 130 m a.s.l.), where the climate is semi-arid Mediterranean. The mean annual temperature at the location is 15.2 °C and the total annual precipitation is 589 mm. The soil is silty loam with a pH 7.6. The experiment was established in autumn 1995. The studied species were *Medicago sativa* L. cv. Ylikí (alfalfa), bred by the Forage Crops and Pastures Institute at Larisa, Greece, and *Festuca arundinacea* cv Festorina (tall fescue) introduced from the USA. The experimental design consisted of completely randomized field plots (1 m x 1 m) of pure and mixed (1:1) stands, with four replications. The sowing density of the pure plots was 4.5 g m<sup>-2</sup> for *Festuca arundinacea* and 4.0 g m<sup>-2</sup> for *Medicago sativa*, and in the mixed stand each component was seeded at half the pure stand seeding rate. All plots were irrigated by sprinkler to maintain field capacity. Fertilizer was not applied. The following levels of cutting intensity were also applied: a) one cutting in the measuring season, at the

stage of full maturity (namely uncut, C0); b) cutting at 7 cm above the soil surface (light cut, C7); and c) cutting at 3 cm above the soil surface (heavy cut, C3). The last two cutting intensities were applied when the first inflorescences appeared during spring and early summer, at about 20-day intervals (four to five cuttings in the measuring season). Two more cuttings were applied during the second and third year, one in early spring and one in autumn. The measurements were repeated for four years (1996-1999). The plant material was separated into the two species components of the mixture and was oven-dried at 75 °C for 48 hours. The above-ground dry biomass (AGDB) of each species and the mixture for each year was obtained. After that, the AGDB over the 4-year period was estimated. The mean annual and seasonal percentage contribution of each species to the AGDM of the whole-plot productivity was calculated.

AGDB data were subjected to analysis of variance (ANOVA) using the SPSS program. LSD values were used to compare treatment means ( $\alpha = 0.05$ ).

## Results and discussion

The total AGDB production of the mixture in uncut treatment (C0) was significantly higher ( $\alpha = 0.05$ ) compared to the cut treatments (C7, C3). Light cut tended to give greater yield than heavy cut, but the difference was not significant. In all cutting treatments a year-to-year oscillation of AGDB was observed (Table 1), with a higher value in the second year and a lower value in the final year (1999). The uncut treatment gave higher annual AGDB than the other two cutting treatments in the first three years, but the yield fell in the final year of the experimentation to a value lower than that of the cut treatments. No significant differences were observed between the two cutting treatments. These results could be explained by taking into account the botanical composition of the mixture, as according to Kunelius *et al.* (2006) legumes in mixtures generally contribute to increased DM production. This aspect was not always confirmed from our results.

Table 1. Above ground dry biomass (AGDB) of the mixture per year and the total AGDB of all years, in  $\text{g m}^{-2}$ , under the uncut treatment (C0), the light cut (C7) and the heavy cut (C3).

	1996	1997	1998	1999	Total
C0	975.1a*	2665.0a	1208.0a	427.5a	5275.6a
C7	560.5b	1392.1b	739.7b	711.6b	3403.9b
C3	549.1b	1206.8b	670.9b	541.5a	2968.4b

\*Columns with different letters within years are significantly different at  $\alpha = 0.05$ .

The mean botanical composition of the mixture, based on the component AGDM yield for the four consecutive years of the experimentation, is presented in Figure 1. According to this, the year-by-year oscillation of the proportions of the two species revealed the effect of temporal climate variability. Generally, the proportion of tall fescue in the mixture was higher than the alfalfa proportion. Lazaridou and Vrahnakis (2005) reported better tall fescue performance in a mixture compared with in its monoculture, probably because the grass takes advantage of the ability of alfalfa to introduce nitrogen into the system. This result revealed differences in the competitive ability of two species. The cutting intensity affected the species' competitive ability as well. In the uncut treatment the tall fescue dominance of the first year changed gradually to dominance of alfalfa in the final year. In contrast, in the cutting treatments the grass proportion remained high throughout the experiment. Light cutting favoured the grass more than heavy cutting.

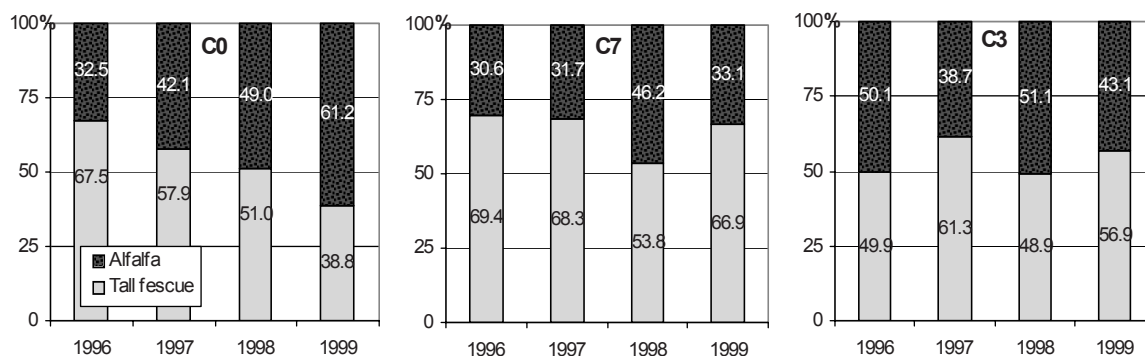


Figure 1. Year-by-year changes of the relative proportions of above-ground dry biomass of alfalfa and tall fescue components of a binary mixture, under three cutting treatments: uncut (C0), light cut (C7) and heavy cut (C3).

The mean seasonal changes of the botanical composition of the mixture over the four years appear in Figure 2. It is well known that grasses perform better in early spring while legumes later. Our results confirm this aspect. In the uncut treatment the proportion of grass was high when the plants were cut in April or June. When cutting was applied in this period the plants had grown under temperatures that favour grass growth. But the proportion of grass was low when the plants were cut in July because this period favours legumes. In the two cutting treatments the proportion pattern was the same, showing a better performance of grass in April than the following months. There were no differences between the two cutting intensities concerning the seasonal effect on plant competitive ability.

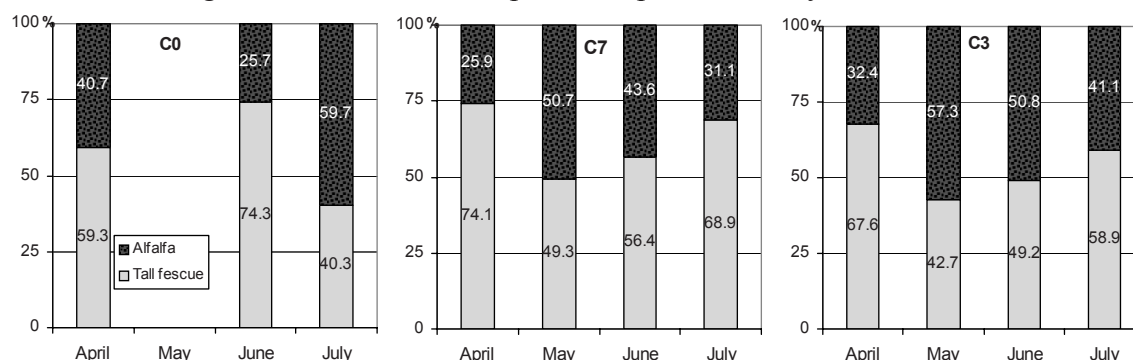


Figure 2. Mean seasonal changes of the proportional composition of alfalfa and tall fescue, components of a binary mixture, under three cutting treatments: uncut (C0), light cut (C7) and heavy cut (C3).

## Conclusions

Increased cutting intensity decreases the annual and the total AGDB of the mixed swards of tall fescue and alfalfa. The botanical composition of the mixtures oscillated within and between growing seasons. The cutting improved the competitive ability of tall fescue, which successfully competed with alfalfa in this Mediterranean climate with irrigation.

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# Productivity and competitive ability evaluation of selected perennial ryegrass varieties, tested in different soil-climatic conditions

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## Abstract

Forage productivity, persistence, disease resistance and competitive ability of selected perennial ryegrass (*Lolium perenne*) varieties were investigated in small-plot trials under contrasting soil and climatic conditions at different locations in the Czech Republic. The productivity of varieties was tested at three different sites: Troubsko (270 m a.s.l.), Zubří (356 m.a.s.l.) and Vatín (540 m a.s.l.). Ryegrass varieties were tested in monoculture and in 50:50 legume/grass mixtures. Five harvests were carried out each year for two years. Results were analysed by ANOVA. The best results were achieved by the diploid cultivar AberElan and tetraploid cultivar Kentaur. Other cultivars responded differently to different soil-climatic conditions. The best varieties in mixtures were achieved with diploid cultivars AberElan, AberDart, Olaf and the tetraploid cultivars Kertak, Jaspis and Alligator. The varieties Kentaur, Kertak, Sirius and Alligator showed the best tolerance of grey snow mould (*Typhula incarnata*).

Keywords: perennial ryegrass, variety, productivity, diseases

## Introduction

In the Czech Republic, perennial ryegrass is often used for the establishment or oversowing of highly productive pastures (Kohoutek *et al.*, 2005). The European Common Catalogue of Varieties of Agricultural Crop Species has been effective in the Czech Republic since the entry of the Czech Republic into the European Union in 2004. It is theoretically possible to sow all varieties included in the catalogue. Unfortunately, the end-use properties of most of the varieties in the European Common Catalogue have not been properly tested in our soil and climatic conditions and so there is a risk that farmers will use cheap seeds of varieties which will not have adequate performance, disease resistance and persistence. The purpose of this paper is to provide information about these traits in selected perennial ryegrass varieties tested under different soil and climatic conditions in the Czech Republic.

## Material and methods

In 2004 a small-plot trial with selected varieties of perennial ryegrass (10 diploid and 10 tetraploid) was established at three different sites: Zubří, Troubsko and Vatín (Table 1).

Table 1. Major characteristics of experimental sites.

Locality	Altitude	Average temperature	Annual rainfall	Soil type
Troubsko	270 m	8.6 °C	540 mm	Luvisol
Zubří	356 m	7.5 °C	886 mm	Cambisol
Vatín	540 m	6.1 °C	736 mm	Acid Cambisol

The varieties were tested in monoculture and in clover-grass mixtures (50% perennial ryegrass, 10% meadow fescue, 10% red fescue, 10% timothy, 10% KBG, 5% red clover, 5% alsike clover). The size of the plots was 10 m<sup>2</sup> and all treatments had three replications. The fertilizer regime consisted of phosphoric and potash fertilizers applied in autumn according to the soil nutrient status. Nitrogen was applied in spring at a rate of 60 kg N ha<sup>-1</sup>; after the first cut at 40 kg N ha<sup>-1</sup> and after cuts two, three and four at 30 kg N ha<sup>-1</sup>. The trial was harvested in five cuts: the first cut was taken when ryegrass had reached a height of 20-25 cm, the second cut was 21 days later, the third cut was 28 days later, the fourth cut 35 days later and the fifth cut was in late September at the latest. The following characteristics were determined: fresh and dry forage yield, proportion of weeds and empty spaces and disease occurrence. In mixtures the weight proportion of varieties in the stand was determined before the first harvest. The results were statistically tested using ANOVA.

## Results and discussion

Preliminary results from two harvest years revealed considerable differences in the forage yield of individual varieties at different sites. Of the diploid varieties very good results were provided by the variety AberElan on all sites. Amongst tetraploid varieties the best was the variety Kentaur. The highest DM yields in monoculture were produced by the diploid varieties AberElan and Olaf in Zubří, by the varieties Mara, Bravo and Aberelan in Troubsko, and by the varieties Premium, Aberelan and Respect in Vatin. Of the tetraploid varieties the highest DM yields were provided by Mustang, Sirius and Kentaur in Zubří, by Alligator and Jantar in Troubsko, and by Kentaur, Baristra and Alligator in Vatin. The yields of the varieties on the different sites are shown in Tables 2-3.

Table 2. Dry matter yield (t ha<sup>-1</sup>) of diploid ryegrass varieties at different experimental sites.

Locality	Troubsko				Zubří				Vatin			
variety	2005	2006	Sum	Stat.	2005	2006	Sum	Stat.	2005	2006	Sum	Stat.
Algol	5.8	4.3	10.1	bc	11.3	8.2	19.5	Ab	8.3	7.1	15.4	E
Olaf	7.2	4.7	11.9	abc	11.9	10.0	21.9	A	8.5	7.1	15.6	de
Mara	8.5	7.5	16.0	A	10.3	7.2	17.5	Bc	9.3	7.8	17.1	bcde
Talon	9.0	3.2	12.2	abc	10.4	9.1	19.5	Ab	8.9	7.8	16.7	cde
Bravo	9.4	5.4	14.8	A	8.9	6.4	15.3	C	9.5	7.7	17.2	bcde
AberElan	7.9	6.2	14.1	ab	10.5	12.1	22.6	A	10.7	9.5	20.2	ab
AberDart	6.4	5.7	12.1	abc	9.8	10.3	20.1	Ab	11.0	7.7	18.7	abcde
Premium	6.5	7.2	13.7	abc	10.8	9.5	20.3	Ab	12.1	9.1	21.2	A
Respect	6.8	5.3	12.1	abc	10.2	10.1	20.3	Ab	10.3	9.0	19.3	abc
Sponsor	6.9	2.8	9.7	C	10.1	10.0	20.1	Ab	10.4	8.4	18.8	abcd
Mean t ha <sup>-1</sup>	7.4	5.2	12.5		10.4	9.3	19.7		9.9	8.1	18.0	

Table 3. Dry matter yield (t ha<sup>-1</sup>) of tetraploid ryegrass varieties at different experimental sites.

Locality	Troubsko				Zubří				Vatin			
Variety	2005	2006	Sum	Stat.	2005	2006	Sum	Stat.	2005	2006	Sum	Stat.
Jantar	7.8	7.0	14.8	A	10.3	8.2	18.5	De	9.0	7.9	16.9	a
Jaspis	5.7	7.7	13.4	A	10.0	7.8	17.8	E	8.9	7.6	16.5	a
Lonar	6.7	4.4	11.1	A	12.2	7.6	19.8	Bcde	9.3	7.7	17	a
Kentaur	6.0	6.7	12.7	A	13.4	8.0	21.4	Abc	10.1	8.6	18.7	a
Kertak	5.4	6.3	11.7	A	11.5	8.7	20.2	Abcde	9.6	7.2	16.8	a
Mustang	6.8	5.9	12.7	A	12.5	10.0	22.5	A	8.7	7.6	16.3	a
Tarpan	6.4	4.6	11.0	A	11.9	9.0	20.9	Abcd	8.6	7.7	16.3	a
Baristra	8.6	5.2	13.8	A	11.3	8.5	19.8	Bcde	9.9	8.3	18.2	a
Sirius	6.9	5.4	12.3	A	11.5	10.5	22	Ab	8.2	8.6	16.8	a
Alligator	8.4	7.1	15.5	A	10.6	8.3	18.9	Cde	9.3	8.5	17.8	a
Mean t ha <sup>-1</sup>	6.9	6.0	12.9		11.5	8.7	20.2		9.9	8.1	18.0	

Major differences in the performance of varieties in mixtures were only observed in the second harvest year. In mixtures, in general, the best performing varieties were the diploid varieties AberElan, AberDart and Olaf and the tetraploid varieties Kertak, Jaspis and Alligator (Table 4).

Table 4. Weight ratio of perennial ryegrass varieties on grass-clover mixture yield.

Ploidy	Variety	Site	1st harvest year				2nd harvest year			
			T	Z	V	mean.	T	Z	V	mean
2n	Algol		71	88	73	78	13	42	48	35
	Olaf		85	94	76	85	19	77	54	50
	Mara		74	92	74	80	17	48	68	45
	Talon		88	99	80	89	14	44	68	42
	Bravo		71	86	83	80	14	34	71	40
	Aberelan		61	97	84	81	16	68	77	54
	Aberdart		78	91	79	83	12	69	74	52
	Premium		56	93	78	76	14	71	58	47
	Respect		80	92	78	84	16	53	75	48
	Sponsor		78	86	82	82	10	36	59	35
4n	Jantar		79	83	81	81	15	43	71	43
	Jaspis		78	97	78	84	13	68	62	47
	Lonar		77	96	77	83	14	52	59	41
	Kentaur		75	96	88	87	13	51	72	45
	Kertak		80	96	78	85	15	85	46	49
	Mustang		87	88	86	87	12	53	55	40
	Tarpan		84	89	70	81	13	54	34	34
	Baristra		67	82	67	72	16	40	44	33
	Sirius		71	97	86	85	16	31	60	35
	Alligator		83	92	79	85	15	69	58	47

Sites: T – Troubsko, Z – Zubří, V – Vatin.

The health of varieties of perennial ryegrass was tested predominantly by long-term snow cover and subsequent infection with grey snow mould. The varieties most infected by this disease were Bravo, Premium and Respekt. These varieties, alone amongst the tested cultivars, were also infected with fungi of the genus *Neotyphodium*. In summer and in autumn ryegrass varieties were also infected with leaf blotches which also infected the above mentioned varieties most (data not shown).

## Conclusion

The results of assessment of productive and non-productive properties of varieties of perennial ryegrass grown in monoculture and in clover-grass mixtures revealed their yield potential in different soil and climatic conditions. The best results were achieved from the diploid cultivar AberElan and tetraploid cultivar Kentaur. Other cultivars showed different productivities in different soil-climatic conditions.

## Acknowledgement

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# Sward regeneration after winter grazing depending on the intensity of sward damage

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## Abstract

Grazing during winter causes considerable sward damage if the grazing management is inadequate. The identification of the maximum extent of tolerable sward damage after winter grazing was the main objective of the project. Prerequisites for tolerable extents of damage were the ability of swards to regenerate in spring or early summer without any intervention by the farmer. Five pastures have been selected from five farms with longstanding practice of year-round outdoor keeping of suckler cows. Plots with different extents of damage after winter grazing had been established in these winter pastures for monitoring grassland regeneration. Differences in botanical composition and percentage of gaps were estimated in monthly intervals. Pasture areas with damage to an extent of 25, 50 and 75% after winter grazing regenerate rapidly in spring and early summer. Completely destroyed swards showed significantly longer periods for regeneration. Concerning the occurrence of therophytes in the swards, no significant difference was evident between damaged and intact swards, even if the plots with an extent of 75% of damaged vegetation cover was considered. Only plots with completely destroyed vegetation (= 100%) showed a significant higher percentage of therophytes and also plant species with a high treading tolerance.

Keywords: winter grazing, vegetation gaps, spring regrowth, botanical composition

## Introduction

Year-round outdoor keeping of cattle can be an efficient and environmentally friendly way of low-input farming even in regions with harsh winter conditions if the specific grazing practice is suitable to avoid excessive sward damage. The intensity of sward damage is related to the intensity of treading and the duration of stay of the livestock. The identification of the maximum extent of tolerable sward damage after winter grazing was the main objective of the project. Prerequisites for tolerable extents of damage were the ability of swards to regenerate in spring or early summer without any intervention by the farmer (e.g. repair sowing).

## Material and methods

Five *Lolio-Cynosuretum* pastures had been selected from five farms with longstanding practice of year-round outdoor keeping of suckler cows and beef cattle with different management strategies. These farms were specifically chosen to obtain monitoring plots. The farms were in the locality of Westerwald highlands, a region between Siegen, Koblenz and Dillenburg, in middle-western Germany. Plots (= 4 m<sup>2</sup>) with different extents of sward damage after winter grazing (= 0, 25, 50, 75 and 100%) had been established in these winter pastures for the monitoring of grassland regeneration. Each level contained four replications per location. The plots were fixed by GPS data and wooden markers in soil. Differences in botanical composition (estimation of yield proportions according to Klapp (1929)), and percentage of gaps (visual estimation) were documented at monthly intervals in spring and summer in the years 2006 and 2007. Plant species were organized in functional groups according different biological attributes, such as nitrogen index (N-value), treading tolerance and life-form (Ellenberg *et al.*, 2001). According to Ellenberg *et al.* (2001), the N-value 1

stands for indicator species for low soil nutrient level, and N-value 9 stands for indicator species for extremely high nutrient level in the soil. In the following, plant species with an N-value between 7 and 9 are assessed to be indicators for a high nitrogen level in soil. Time series data have been checked with repeated measures ANOVA by using software SPSS 12.0.

## Results

Pasture areas with 25, 50 and 75% of damage after winter pasturing regenerate themselves rapidly between spring and early summer. Completely damaged swards need significantly longer periods for regeneration. Figure 1 shows the development of mean percentage of gaps of all plots.

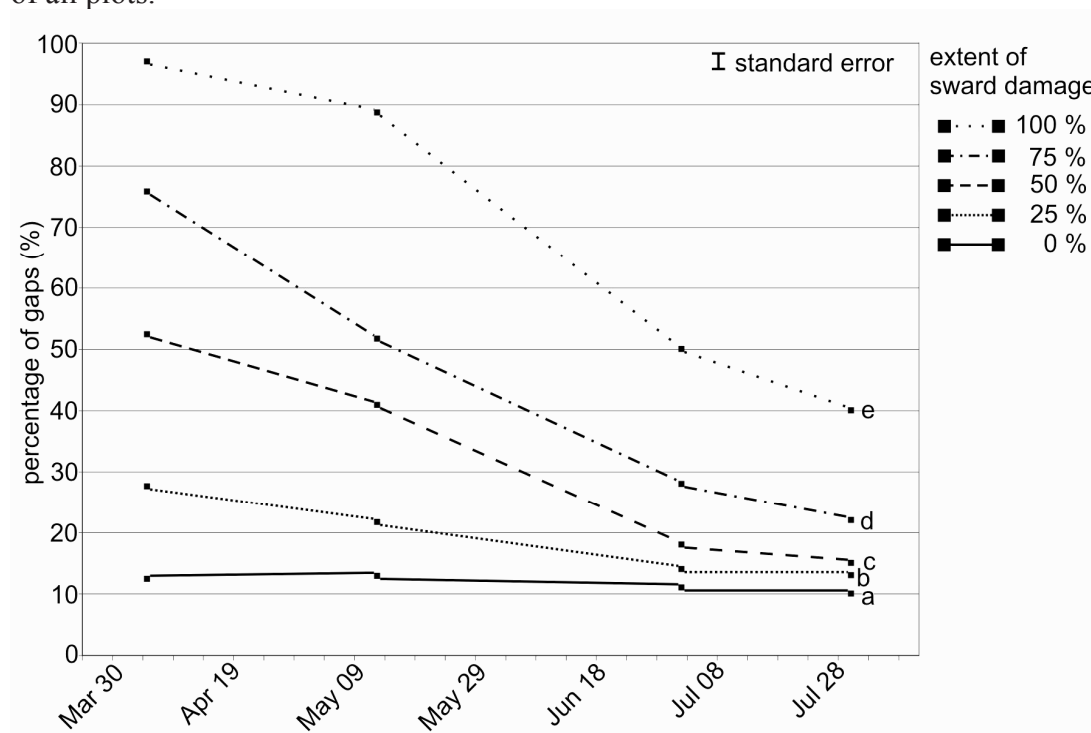


Figure 1. Development of the percentage of gaps (mean value of all plots) in spring and summer 2007 depending on the extent of sward damage after winter pasturing. Significant differences ( $P < 0.05$ ) are shown in lowercase letters.

According to percentage of therophytes (see Ellenberg *et al.*, 2001) swards with an extent of damage up to 75% of total plot size do not differ from unharmed plots. Only plots with completely destroyed vegetation show a significantly higher percentage of therophytes and plant species with high treading tolerance (Figure 2). Also, the yield proportions of nitrogen indicator species (N-value 7-9) show a higher percentage on destroyed plots.

## Discussion

Because of higher growth rates in spring, the progress of regeneration of sward damage after winter grazing is faster than the progress in late summer or autumn after regular summer grazing. For regeneration without any intervention by farmers (e.g. repair sowing) it is necessary to ensure a certain percentage of remaining vegetation.

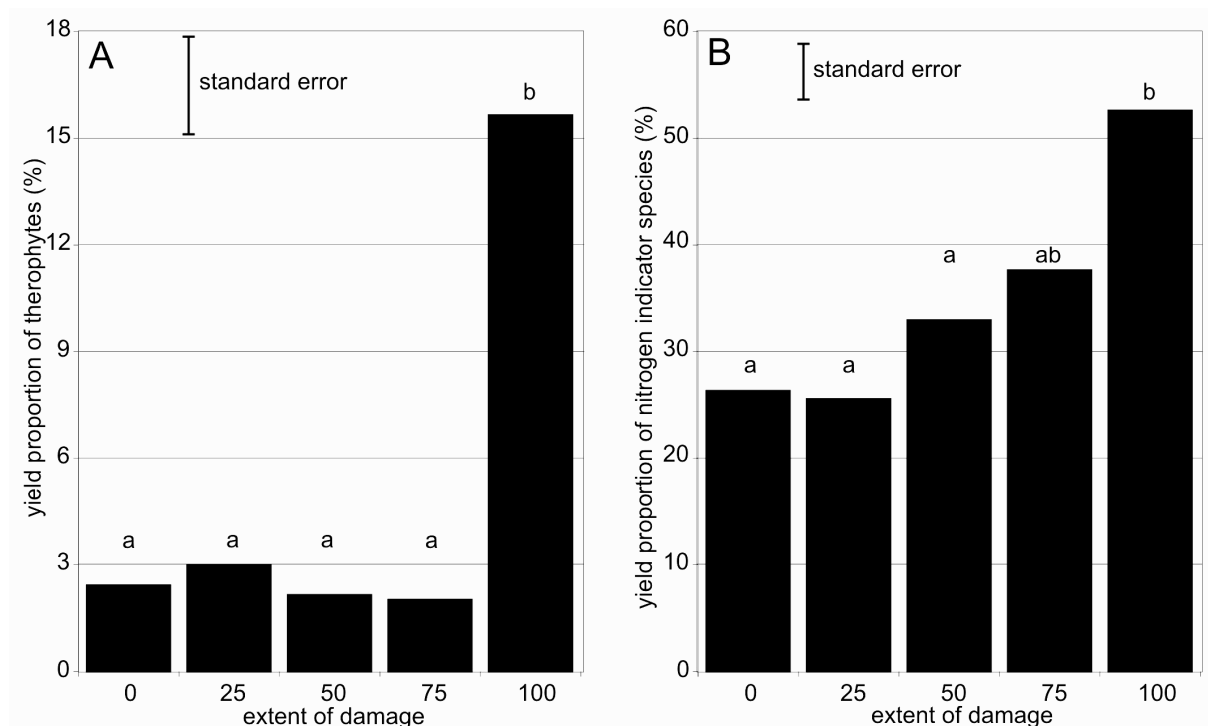


Figure 2. Mean yield proportion of therophytes (A) and nitrogen indicator species (B) in May 2007 depending on the extent of sward damage after winter pasturing. Significant differences ( $P < 0.05$ ) are shown in lowercase letters.

If the distribution of remaining perennial plant individuals is relatively homogeneous, sward damages up to 75% are tolerable. Most completely damaged areas are found close to feeding grounds or resting places. In these areas the closing of gaps is much harder and the number of therophytes is significantly increased. These species, which can not establish in a closed sward, find better conditions for germination and establishment in areas with the highest disturbance intensity. Indeed, the therophyte species contribute to close the gaps but they decrease the yield and forage quality. The higher rate of nitrogen indicator species in destroyed swards is caused by a higher nitrogen entry in soil around feeding grounds as documented by Opitz v. Boberfeld *et al.* (2004): With increasing distance from feeding grounds the nitrogen entry in soil and also the damage of sward by treading decrease.

## Conclusions

It is advisable to allow winter grazing on pastures with larger extensions to obtain locally and diffusely distributed areas of damages instead of restricting winter grazing areas to small pastures with high stock densities. It is necessary to avoid areas with nearly completely damaged vegetation in spring. This is possible by frequent rotation, limited stock densities and the use of mobile feeding and watering equipment.

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# Weed flora dynamics during the first years of grassland establishment

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## Abstract

A long term experiment was initiated in 2005 at the INRA research centre of Lusignan (western France) to evaluate environmental impacts of temporary grasslands. One of the objectives is to study the dynamics of plant community, seeded species as well as weed flora, taking into account grassland duration in crop rotation and nitrogen supply. Weeds originate from the seed bank and seed dispersion. Seed bank was evaluated at the beginning of the experiment and aboveground weed flora was observed once a year. Results show a modification of the weed community structure during the first years of grassland establishment. Quantitatively, with a decrease of specific richness and Shannon index, and qualitatively, with a reduction of cool-growing plants that were very important in the first years to the benefit of perennial species that are frequently observed in grasslands. Weed flora in low N treatment differed from high N treatment in the third year.

Keywords: temporary grassland, weeds, plant diversity, N fertilization, long term experiment

## Introduction

Grasslands, which represent a large part of land use in France, have seen their surface area decline by 28% since 1950 (French agriculture statistical data). Numerous studies have shown the effects of grasslands on several ecological processes or functions (nutrient circulation, carbon sequestration, biodiversity support) (e.g. Soussana *et al.* (2004) for C sequestration). Grasslands may contribute to habitat quality for biodiversity in several ways: protection, reproduction and trophic resources availability (Wolff *et al.*, 2001). The long term experiment ORE – ACBB (Observatory Environmental Research, Agro-ecosystems, Biogeochemical Cycles, and Biodiversity) was initiated in 2005 at the INRA research centre of Lusignan (western France) to evaluate environmental impacts of grasslands (Lemaire *et al.*, 2005). As well as permanent grasslands, temporary grasslands may play an important role in ecological functions but also in agronomic issues by modifying seed bank qualitatively and/or quantitatively (Bellinder *et al.*, 2004) and so affecting weed populations remaining in the following crops. We hypothesized that both functions, ecological and agronomical, depend on plant community structure and dynamics in temporary grasslands. We focused the analysis on weed flora considering the impact of grassland management, i.e. grassland duration in crop rotation and nitrogen supply. We presented results of aboveground observations in temporary grasslands from 2005 to 2007, i.e. during their first years of establishment. Seed bank evolution is also studied in this work as it may contribute to plant community dynamics, but it is not presented here.

## Materials and methods

The experiment takes place at the INRA research centre of Lusignan (western France). The experimental design is composed of four blocks in which five treatments were randomly allocated. Each plot is 0.4 ha. The treatments are differentiated by duration (0, 3, 6 and 20 years) and N fertilization (with or without) of grasslands as followed. T1: three-year rotation of annual crops without grassland (maize in 2005, wheat in 2006, barley in 2007, and so on);

T2: three years of annual crops (maize in 2005, wheat in 2006, barley in 2007) followed by three years of N-supplied grassland; T3: six years of N-supplied grassland followed by three years of annual crops (grassland from 2005 to 2010); T4: six years of grassland without N followed by three years of annual crops (grassland from 2005 to 2010); T5: permanent grassland with N supply. Grasslands were seeded in May 2005 with a mixture of *Lolium perenne*, *Festuca arundinacea* and *Dactylis glomerata*. Aboveground weeds were observed at least once a year, during spring. Only treatments T1, T2 and T5 were observed in the first year, and then all treatments were surveyed. Plants were observed in a 0.25 m<sup>2</sup> circle at 13 locations along a cross path delineated in each plot. Weed species were noted using a modified Barralis method. Species richness (S) and abundance (A), and diversity indices, namely Shannon (H) and evenness (E) indices, were computed from the floristic data (Légère *et al.*, 2005). Abundance per plot is calculated by adding up the Barralis notes. Analyses of variance were performed with R freeware using lm and anova functions.

## Results and discussion

Only results on grasslands are presented, i.e. T3, T4 and T5 treatments. At the beginning of the experiment (observations in 2005), there are no significant effects of block and treatment (T1, T2 and T5) on S, H and E. A is significantly higher in blocks 1 and 2 than in blocks 3 and 4 ( $P$  value = 0.027), likely due to a different cultural past. For years 2006 and 2007, effects of treatments (T3, T4, T5) and year x treatment interaction are not significant. Variables A, S and H, excepted E, are significantly decreasing with years ( $P < 0.001$ ). Mean species richness per plot decreases from  $14.8 \pm 2.6$  in 2005 to  $4.8 \pm 1.6$  in 2007 and mean abundance from  $130 \pm 40$  to  $26 \pm 19$ . This mainly explains the Shannon index decline that depends on both variables. Evenness index remains stable around  $0.8 \pm 0.07$  (Figure 1). This relatively high value indicates a quite homogenous distribution of species abundance in plots. We point out for the T4 treatment (without N), higher values of S and H in 2006 and 2007, and of E in 2006, even though not significant.

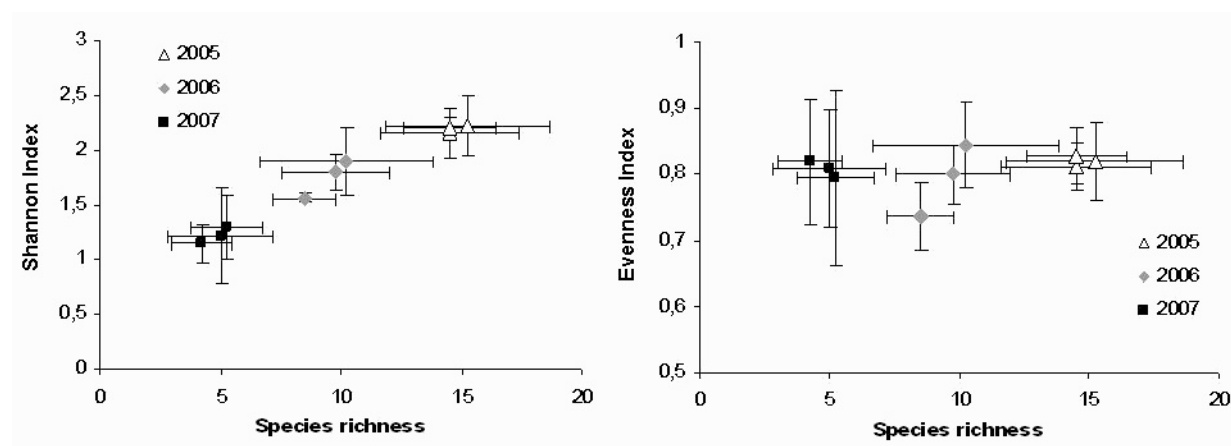


Figure 1. Shannon (on the left) and evenness (on the right) indices plotted against species richness in grasslands in 2006 and 2007. Data obtained at the beginning of the experiment (2005) are presented for information. Bars are standard deviations.

The total number of weed species over all treatments show a decrease from 31 in 2005 to 16 in 2007 with a modification of plant community. Main species encountered in 2005 are *Polygonum aviculare* (polavi), *Capsella bursa-pastoris* (capbur), *Sonchus asper* (sonasp) and *Solanum nigrum* (solnig). These are annual plants with long flowering period except for soling, which flowers in summer. Solnig was not observed in the following years, possibly



because observations were performed sooner (25 May 2005, 27 April 2006 and 2 April 2007). Polavi and capbur were still present in 2006 with a high abundance but no more observed in 2007. Sonasp abundance greatly decreased in 2006 and was almost nil in 2007. Other common weed species (*Polygonum persicaria*, *Poa annua*, *Chenopodium album*, for instance) present the same trend but with lower abundance. *Veronica persicaria* and *Poa trivialis* remained stable through the period, the first being an annual species and the second a perennial one. Some species, mainly perennial or biannual, see their abundance increased: by a lot for *Taraxacum officinalis*, but much less for *Hypericum perforatum* and *Picris echioides*. The abundance of one annual plant, *Cerastium glomeratum*, increases.

Considering T4 treatment in 2007, we observed the presence of one particular species *Aphanes arvensis*, not observed in the other treatments. Some species, such as *Trifolium repens*, present higher abundance in the low N treatment.

Diversity indices measured in this study are within the range of those reported for similar cropping systems (Légère *et al.*, 2005). Shannon index in weed community is commonly less than 2. Evenness index is slightly higher than those found in previous studies, around 0.6, presumably due to the estimation of abundance with the modified Barralis notation. A large decline of almost all diversity variables, species richness and abundance and Shannon index, in the first years of grassland is related to the introduction of seeded species. They colonized the environment and directly competed with weeds for light and nutrients. Higher diversity found in low N treatment may indicate a differential response to N deficiency of weed community and/or seeded species. These later would grow slower or lesser than in N-supplied treatments to the benefit of weeds. Data on the proportion of bare soil and seeded species growth have been monitored and it would be interesting to include them in this study.

Weed species at the beginning of the experiment are commonly found in cultures. They are mainly annual dicotyledons. A slight change in plant community is observed during the first three years to the advantage of perennial species, which are more frequent in grasslands. The third year, low N grasslands slightly differ in their plant community, in particular with the presence of *Trifolium repens*. This species, which is a legume, is less dependant on N availability in the soil. This can explain its higher abundance in unfertilized grassland.

To complete this study, a functional approach of species groups relevant for agronomic or environmental purposes is in progress.

## Acknowledgments

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# Forage yields in urban populations of hairy vetch (*Vicia villosa* Roth) from Serbia

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## Abstract

Hairy vetch (*Vicia villosa* Roth) is widely distributed in Serbia and often grows in diverse environments, including urban areas. A small-plot trial was carried out in 2006 and 2007 at the Experimental Field of the Institute of Field and Vegetable Crops at Rimski Šančevi. It included seven hairy vetch populations collected from several urban regions of Belgrade and Novi Sad. All seven populations were sown in early October, at a rate of 180 viable seeds m<sup>-2</sup>, and cut at the stages of full flowering and formation of the first pods. With an average crop density before cutting of 150 plants m<sup>-2</sup>, the populations MM 04/13 and MM 04/18 proved to be the most winter hardy. The average values of plant height ranged from 100 cm in MM/04/20 to 235 cm in MM 03/26. The population MM 03/26 had the greatest numbers of both stems and lateral branches (15.0 plant<sup>-1</sup>) and internodes (163.0 plant<sup>-1</sup>). The highest green forage and forage dry matter yields were in the population MM 04/19 (50.9 Mg ha<sup>-1</sup> and 11.7 Mg ha<sup>-1</sup>). The highest proportion of forage dry matter was in MM 04/09 (0.33).

Keywords: hairy vetch, urban populations, green forage yield, forage dry matter yield, forage yield components

## Introduction

Hairy vetch (*Vicia villosa* Roth) is a traditional annual forage legume in both Serbia and the Balkans, along with common (*Vicia sativa* L.) and Hungarian (*Vicia pannonica* Crantz) vetches (Mikić *et al.*, 2006). It is grown as an autumn-sown crop and considered one of the most resistant cultivated species to low temperatures (Vučković, 1999).

Hairy vetch is often found growing together with large-flowered vetch (*Vicia grandiflora* Scop.) and narrow-leaved vetch (*Vicia sativa* subsp. *nigra* (L.) Ehrh.) in diverse environments, including urban areas (Ćupina *et al.*, 2007). The Annual Forage Legumes Collection (AFLCNS) of the Institute of Field and Vegetable Crops (IFVCNS) is constantly enriched with the local landraces and wild populations of Serbian origin, mostly collected in the region of Novi Sad (Tomić *et al.*, 2005).

The aim of the study was to evaluate forage yield of seven wild Serbian urban populations of hairy vetch, collected from several sites in the urban areas of Belgrade and Novi Sad.

## Materials and methods

A small-plot trial was carried out in 2006 and 2007 at the Experimental Field of the IFVCNS at Rimski Šančevi. It included seven hairy vetch populations collected in the urban regions of Belgrade (MM 03/05 and MM 03/26) and Novi Sad (MM 04/09, MM 04/13, MM 04/18, MM 04/19 and MM 04/20) during the expeditions that the IFVCNS undertook in 2003 and 2004. The collected populations were included in the AFLCNS, maintained in spatial isolation in order to avoid cross-pollination between populations and multiplied with the main aim of the evaluation of their agronomic characteristics related to forage.

All seven populations were sown in early October, at a rate of 180 viable seeds m<sup>-2</sup>, a plot size of 5 m<sup>2</sup> and three replicates. All populations were also cut in the stages of full flowering and formation of the first pods, as an optimal balance between forage yield and quality (Mejakić and Nedović, 1996), that was in the second half of June in both years.

Number of plants before cutting (m<sup>-2</sup>) was determined at the beginning of flowering, with overwintering determined as a ratio between the plant numbers in early winter (i.e. early December), and early spring (early March). Plant height (cm), number of stems and lateral branches (plant<sup>-1</sup>), number of internodes (plant<sup>-1</sup>) and green forage yield per plant (g) were determined from plant samples taken just before cutting. Green forage yield per area unit (Mg ha<sup>-1</sup>) was measured *in situ* immediately after cutting. Forage dry matter yield (Mg ha<sup>-1</sup>) was calculated on the basis of the values of green forage yields and the values of forage dry matter proportion, with the latter being a ratio of forage samples mass after and before the drying at room temperature, and having taken in account a dry matter content of 85%.

Results were processed by analysis of variance (ANOVA) applying the Least Significant Difference (LSD) test using the computer software MSTAT-C.

## Results and discussion

Table 1. Average values of forage yield components in seven populations of hairy vetch for the years of 2006 and 2007 at Rimski Šančevi.

Population	Number of plants (m <sup>-2</sup> )	Plant height (cm)	Number of stems and lateral branches (plant <sup>-1</sup> )	Number of internodes (plant <sup>-1</sup> )
MM 03/05	90	190	2.0	58.0
MM 03/26	90	235	15.0	163.0
MM 04/09	135	120	4.2	63.4
MM 04/13	150	125	5.4	73.2
MM 04/18	150	120	6.1	100.9
MM 04/19	136	145	8.3	110.2
MM 04/20	133	100	8.1	140.3
LSD <sub>0.05</sub>	15	14	2.6	21.4
LSD <sub>0.01</sub>	21	19	4.0	35.7

Table 2. Average values of forage yields in seven populations of hairy vetch for the years of 2006 and 2007 at Rimski Šančevi.

Population	Green forage yield (g plant <sup>-1</sup> )	Green forage yield (Mg ha <sup>-1</sup> )	Forage dry matter yield (g plant <sup>-1</sup> )	Forage dry matter yield (Mg ha <sup>-1</sup> )	Forage dry matter proportion
MM 03/05	36.89	33.2	10.71	9.6	0.29
MM 03/26	52.60	47.3	10.69	9.5	0.20
MM 04/19	17.26	23.3	5.70	7.7	0.33
MM 04/13	19.58	29.4	4.00	6.0	0.20
MM 04/18	30.43	45.6	6.93	10.4	0.23
MM 04/19	37.68	50.9	8.70	11.7	0.23
MM 04/20	29.57	39.9	5.70	7.7	0.19
LSD <sub>0.05</sub>	6.49	5.6	1.72	1.3	0.02
LSD <sub>0.01</sub>	8.04	7.8	2.23	1.9	0.03

There were significant differences at the levels of both 0.05 and 0.01 in all monitored characteristics between the examined urban populations of hairy vetch (Table 1).

With an average crop density before cutting of 150 plants m<sup>-2</sup>, the populations MM 04/13 and MM 04/18 proved as the most winter hardy, while the populations MM 03/05 and MM 03/26, with an average crop density before cutting of 90 plants m<sup>-2</sup>, were the most susceptible to low temperatures. The average values of plant height, ranging from 100 cm in MM/04/20 to 235 cm in MM 03/26, may be considered as within the average of the species (Erić *et al.*,

1996). The population MM 03/26 had the greatest numbers of both stems and lateral branches ( $15.0 \text{ plant}^{-1}$ ) and internodes ( $163.0 \text{ plant}^{-1}$ ), while the population MM 03/05 had the smallest numbers of both stems and lateral branches ( $2.0 \text{ plant}^{-1}$ ) and internodes ( $58.0 \text{ plant}^{-1}$ ).

The average green forage yield per plant varied between 17.26 g in MM 04/19 and 52.60 g in MM 03/26, while the average forage dry matter yield per plant varied between 4.00 g in MM 04/13 and 10.71 g in MM 03/05 (Table 2). The population MM 04/19 had the highest yields per area unit of both green forage and forage dry matter ( $50.9 \text{ Mg ha}^{-1}$  and  $11.7 \text{ Mg ha}^{-1}$ ), proving that wild hairy vetch populations can be regarded as with high potential in comparison to hairy vetch advanced cultivars (Mihailović *et al.*, 2006a). The lowest forage yields were in MM 04/09 with  $23.3 \text{ Mg ha}^{-1}$  of green forage and MM 04/13 with  $4.0 \text{ Mg ha}^{-1}$  of dry matter. The highest proportion of forage dry matter was in MM 04/09 (0.33), while the lowest proportion of forage dry matter was in MM 04/20 (0.19).

## Conclusions

The wild populations of hairy vetch have shown a high level of variability of agronomic characteristics and may be considered as having a considerable potential for its utilization in breeding and the development of new cultivars for forage production and green manure, where green forage yield plays more important role. The future research should focus on quality, especially crude and digestible protein and fibre content, and thus provide more complete results.

## Acknowledgements

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## Forage yields in urban populations of narrow-leafed vetch (*Vicia sativa* subsp. *nigra* (L.) Ehrh.) from Serbia

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### Abstract

Narrow-leafed vetch (*Vicia sativa* subsp. *nigra* (L.) Ehrh.) often grows in diverse environments in Serbia, including urban areas. A small-plot trial was carried out in 2006 and 2007 at the Experimental Field of the Institute of Field and Vegetable Crops at Rimski Šančevi. It included ten populations of narrow-leafed vetch collected in several urban regions of Serbia. All ten populations were sown in early October, at a rate of 180 viable seeds m<sup>-2</sup>, and cut at the stages of full flowering and formation of the first pods. The average values of plant height ranged from 30 cm in the populations MM 05/08 and MM 05/09 to 76 cm in the population MM 02/01. The population MM 05/08 has the greatest numbers of stems and lateral branches (20.0 plant<sup>-1</sup>) and internodes (235.0 plant<sup>-1</sup>). The highest green forage and forage dry matter yields were in the population MM 02/02 (33.4 Mg ha<sup>-1</sup> and 7.3 Mg ha<sup>-1</sup>). The average values of forage dry matter proportion varied between 0.19 in the population MM 03/17 and 0.35 in the population MM 03/11.

Keywords: narrow-leafed vetch, urban populations, green forage yield, forage dry matter yield, forage yield components

### Introduction

Common vetch (*Vicia sativa* L.) is considered one of the most important species of the genus *Vicia* L. (Matić *et al.*, 2005). Common vetch comprises five subspecies, namely *amphicarpa* (L.) Batt., *cordata* (Wulfen ex Hoppe) Batt., *macrocarpa* (Moris) Arcang., *nigra* (L.) Ehrh. and *sativa*, with the fourth one, known as narrow-leafed or black-pod vetch, as the most widely distributed of all (Maxted, 1995).

Narrow-leafed vetch is rather widespread all over Serbia, where it is frequently found growing together with large-flowered vetch (*Vicia grandiflora* Scop.) and hairy vetch (*Vicia villosa* Roth) in diverse environments, including urban areas. The majority of such populations germinate during autumn, flower in late April and May and bring pods and seeds in late May and throughout June, with an ability to regenerate after cutting and to produce seed more than once a year. The Annual Forage Legumes Collection (AFLCNS) of the Institute of Field and Vegetable Crops in Novi Sad (IFVCNS) is constantly enriched with the local landraces and wild populations of Serbian origin, mostly collected in the region of Novi Sad and the mountain of Fruška Gora (Tomić *et al.*, 2005).

The aim of the study was to evaluate forage yield of ten wild Serbian urban populations of narrow-leafed vetch, collected from several sites in the urban areas of Belgrade and Novi Sad.

### Materials and methods

A small-plot trial was carried out in 2006 and 2007 at the Experimental Field of the Institute of Field and Vegetable Crops at Rimski Šančevi. It included ten wild narrow-leafed vetch populations collected in the urban regions of Belgrade (MM 02/03, MM 02/04, MM 03/08, MM 03/09 and MM 03/17) and Novi Sad (MM 02/01, MM 02/02, MM 03/11, MM 05/08 and



MM 05/09) during the expeditions that the IFVCNS undertook in 2002, 2003 and 2005. The collected populations were included in the AFLCNS, maintained and multiplied with the main aim of the evaluation of their agronomic characteristics related to forage.

All ten populations were sown in early October, at a rate of 180 viable seeds m<sup>-2</sup>, a plot size of 5 m<sup>2</sup> and three replicates. All populations were also cut in the stages of full flowering and formation of the first pods, as an optimal balance between forage yield and quality (Mihailović *et al.*, 2005), that was in the first half of May in both years.

Plant height (cm), numbers of stems and lateral branches (plant<sup>-1</sup>), number of internodes (plant<sup>-1</sup>) and green forage yield per plant (g) were determined from plant samples taken just before cutting. Green forage yield per area unit (Mg ha<sup>-1</sup>) was measured *in situ* immediately after cutting. Forage dry matter yield (Mg ha<sup>-1</sup>) was calculated on the basis of the values of green forage yields and the values of forage dry matter proportion, with the latter being a ratio of forage samples mass after and before the drying at room temperature, and having taken in account a dry matter content of 85%.

Results were processed by analysis of variance (ANOVA) applying the Least Significant Difference (LSD) test using the computer software MSTAT-C.

## Results and discussion

There were significant differences at both levels of 0.05 and 0.01 in all forage yield components among the ten examined wild populations of narrow-leaved vetch (Table 1).

Table 1. Average values of forage yield components in ten wild populations of narrow-leaved vetch for the years of 2006 and 2007 at Rimski Šančevi.

Population	Plant height (cm)	Number of stems and lateral branches (plant <sup>-1</sup> )	Number of internodes (plant <sup>-1</sup> )
MM 02/01	76	13.9	164.4
MM 02/02	60	8.2	50.0
MM 02/03	42	6.2	67.2
MM 02/04	35	4.0	139.4
MM 03/08	72	5.8	71.2
MM 03/09	70	4.9	84.2
MM 03/11	35	12.3	119.3
MM 03/17	62	7.3	79.3
MM 05/08	30	20.0	235.0
MM 05/09	30	14.5	139.4
LSD <sub>0.05</sub>	6	2.5	24.3
LSD <sub>0.01</sub>	14	3.8	35.8

The average values of plant height ranged from 30 cm in the populations MM 05/08 and MM 05/09 to 76 cm in the population MM 02/01. The population MM 05/08 has the greatest number of stems and lateral branches (20.0 plant<sup>-1</sup>), while the population MM 02/04 had the smallest number of stems and lateral branches (4.0 plant<sup>-1</sup>). The greatest number of internodes was in the population MM 05/08 (235.0 plant<sup>-1</sup>), while the smallest number of internodes was in the population MM 02/02 (50.0 plant<sup>-1</sup>).

The population MM 02/02 had the highest average green forage yields per both plant and area unit (22.27 g plant<sup>-1</sup> and 33.4 Mg ha<sup>-1</sup>), significantly higher in comparison with all other nine examined urban populations of narrow-leaved vetch (Table 2). The same population had the highest forage dry matter yields per both plant and area unit (4.84 g plant<sup>-1</sup> and 7.3 Mg ha<sup>-1</sup>). With significant differences at both levels of 0.05 and 0.01, average values of forage dry matter proportion varied between 0.19 in the population MM 03/17 and 0.35 in the population MM 03/11. It is notable that the highest forage yields were in the populations with moderate

values of all forage yield components, confirming the importance of an optimal relationship between forage yield components for high and stable forage yields.

The ten urban populations of narrow-leafed populations produced lower green forage and forage dry matter yields than the cultivars of common vetch, but had higher forage dry matter proportion (Mihailović *et al.*, 2003).

Table 2. Average values of forage yields in ten wild populations of narrow-leafed vetch for the years of 2006 and 2007 at Rimski Šančevi.

Population	Green forage yield (g plant <sup>-1</sup> )	Green forage yield (Mg ha <sup>-1</sup> )	Forage dry matter yield (g plant <sup>-1</sup> )	Forage dry matter yield (Mg ha <sup>-1</sup> )	Forage dry matter proportion
MM 02/01	12.24	18.4	3.32	5.0	0.27
MM 02/02	22.27	33.4	4.84	7.3	0.22
MM 02/03	9.11	13.7	2.90	4.3	0.32
MM 02/04	9.66	14.5	2.92	4.4	0.30
MM 03/08	11.57	17.4	3.32	5.1	0.29
MM 03/09	10.50	15.8	3.35	5.1	0.32
MM 03/11	9.43	14.1	3.34	5.0	0.35
MM 03/17	15.40	23.1	2.87	4.2	0.19
MM 05/08	14.12	21.2	2.92	4.4	0.21
MM 05/09	13.04	19.6	3.34	5.0	0.26
LSD <sub>0.05</sub>	3.18	4.8	0.78	1.1	0.05
LSD <sub>0.01</sub>	5.03	6.3	1.23	1.7	0.08

## Conclusions

The urban populations of narrow-leafed vetch have shown a high level of variability of the characteristics of agronomic importance and may be regarded as having a considerable potential for the development of the first Serbian narrow-leafed vetch cultivars, suitable for forage production and green manure, where green forage yield is of greater importance. Further evaluation should include forage quality parameters, such as crude and digestible protein content and fibre content, as well as more data on seed yield and issues related to their maintenance.

## Acknowledgements

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# Effects of composted, pelletized and anaerobically digested sewage sludge on pasture production after sowing in a silvopastoral system

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## Abstract

Municipal sewage sludge production has increased in recent years in Europe resulting in increased sewage sludge disposal problems. EU promotes sewage sludge usage in agriculture to promote nutrient recycling. Sewage sludge should be stabilized before use, but stabilization methods affect its fertilizer capacity. This aim of this study was to evaluate the effects of different types of sewage sludge (compost, anaerobic, pelletized) on pasture production under silvopastoral systems. Composted and pelletized sludge gave the best tree and pasture production. Pelletized sludge should be promoted on the basis of storage and transport costs.

## Introduction

Agroforestry systems are a sustainable way of land use that is promoted by the EU rural development policy (EU, 2006). The relatively fast growing *Pinus pinaster* is the main forest species used in reforestation in Galicia. Current silviculture for this species (Rodríguez-Soalleiro *et al.*, 1997) promotes mechanical clearing every 3-4 years, but the high cost means most foresters do not do it. The resultant understorey becomes combustible and Galicia is one of the most fire-prone regions of Europe (Rigueiro *et al.*, 2005). Once trees are established, agroforestry system management should be applied to promote pasture and tree growth. Fertilization is most important to increase pasture production, but fertilization practices can modify the competition between trees and pasture. Options should be applied that enhance tree and pasture growth. The efficiency of organic fertilizers depends on the proportions of fast-released mineral nitrogen (N) (mainly nitrate or ammonium), and slow-release organic N. For this reason, efficiency of slurry is important compared with municipal sewage sludge.

Municipal sewage sludge production has increased in recent years due to the EU regulation which promotes inland water quality. Sewage sludge can be used as fertilizer to promote nutrient recycling and avoid dump disposal. Sewage sludge coming from the purification of municipal waters could be stabilized through anaerobic, aerobic, liming or incineration processes. Anaerobic process is the most important means of stabilizing sewage sludge for big cities due to its high speed of stabilization. However, sewage sludge has high water content (*ca.* 25%) and it is difficult to spread; for this reason some alternatives are proposed, e.g. compost and pelletization, which makes it cheaper to transport and store the product until it is used as fertilizer. Nitrogen efficiency of sewage sludge depends on the processing method.

## Material and methods

The experiment was carried out during 2005 and 2006 in NW Spain (43°14'N, 7°21'W; altitude 465 m a.s.l.) in a *Pinus pinaster* Aiton plantation. Initial water soil pH was around 5.5. Annual precipitation of the area was 1,083 mm (mean), but was 824 and 799 in 2005 and 2006, respectively. Rainfall was reduced during the summer period. Experimental design was randomized block with five treatments: (a) Control 1, which consisted in no fertilization (NF) which is a traditional management for plantations in the area, (b) Control 2 e(Min), which consisted of fertilization during two years with 500 kg ha<sup>-1</sup> of 8:24:16 at the start of the growing season (traditional management for agrarian fields in the area), (c) Sewage sludge



composted (total input of 320 kg N ha<sup>-1</sup> in the first year, applied at establishment), (d) Anaerobically digested sludge (a total input of 320 kg N ha<sup>-1</sup> in the first year of establishment), and (e) dehydrated and pelletized sludge (an input of 320 kg N ha<sup>-1</sup>, applied as 160 kg N ha<sup>-1</sup> in the first year and 80 kg N ha<sup>-1</sup> at the end of each winter). Plantation and sowing were made in winter and at the end of the spring, respectively. Before plantation, land was ploughed aiming to incorporate organic compost and anaerobic-digested sludge. All sewage sludges were within the Spanish regulation for applying residues in agriculture (Royal Decree 1310/90). Experimental plots consisted of a perfect square of 25 meters with 5 x 5 trees. Trees were established at a density of 1,111 trees ha<sup>-1</sup> and occupied 144 m<sup>2</sup> (12 x 12). At the end of the spring and autumn, pasture production was estimated through harvesting four squares each of 0.03 m<sup>2</sup>, which were taken to the laboratory to estimate DM production. ANOVA and LSD test were used for statistical analyses. Treatments, blocks and years and the double interactions were used in the ANOVA analyses as factors.

## Results and discussion

Tree growth (height and diameter) as a result of the treatment effects can be seen in Figure 1. *Pinus pinaster* tree growth was very good at two years after establishment (Castillón-Palomeque *et al.*, 2002). Tree diameter was significantly affected by treatments, anaerobic sewage sludge treatment the worst treatment for tree growth. Negative effect of tree diameter as a result of anaerobic digested sludge application was also found for *Pinus radiata* D. Don in forest areas. However, the application of anaerobic sewage sludge had a positive effect on *Eucalyptus nitens* (H. Deane & Maiden) maiden growth (Mosquera-Losada *et al.*, 2007).

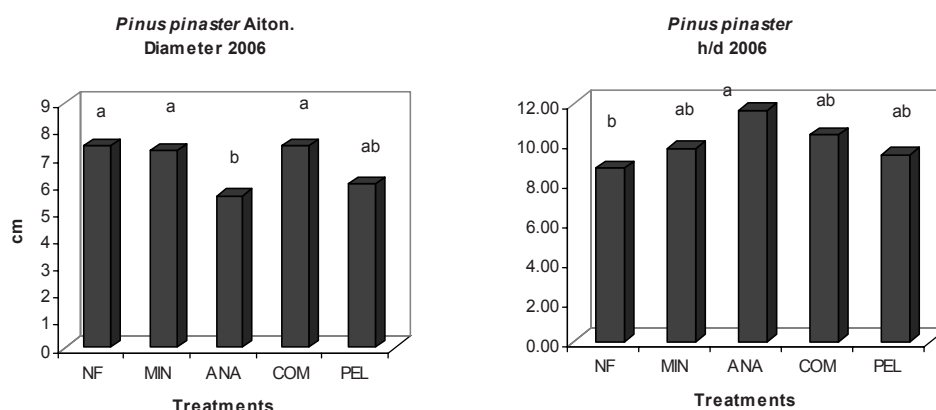


Figure 1. *Pinus pinaster* tree growth (diameter and h/d relationship) for different treatments (NF: no fertilization, MIN: mineral, ANA: anaerobic, COM: compost, PEL: pelletized).

The effect of treatments on annual pasture production is shown in Figure 2. Pasture production was enhanced by compost and pellet application in both years. No-fertilization enhanced tree growth, but not pasture production. However, organic fertilization promoted pasture and tree growth, with the exception of the use of anaerobic-sludge. A similar experiment, on very acid soils in Galicia, demonstrated that anaerobic-sludge reduced *Pinus radiata* D. Don growth, which was explained by the increment of pasture production. However, mineral fertilization increased tree growth due to the low pasture production in the forest soil. Tree growth in very acid soils without fertilization is very small. Tree growth is slower than pasture development, which responds quickly to fertilization. When soil is very poor, organic fertilization enhances pasture growth because it increases pH; however, if only mineral nitrogen is applied no pasture growth is obtained and trees can use the nitrogen. When no fertilization is applied, in spite of the high soil organic matter content which has important

potential for releasing nutrients, tree growth and pasture did not grow because the pH was too low for nutrient release. In the present experiment, soil nutrient release is high enough to benefit tree growth. However, background soil nutrient status could not satisfy pasture requirements and this is limited when no fertilization is applied. On the basis of the present experiment, pellet-stabilized sewage sludge should be promoted, because it has a very low water content, and transport as well as storage is cheaper.

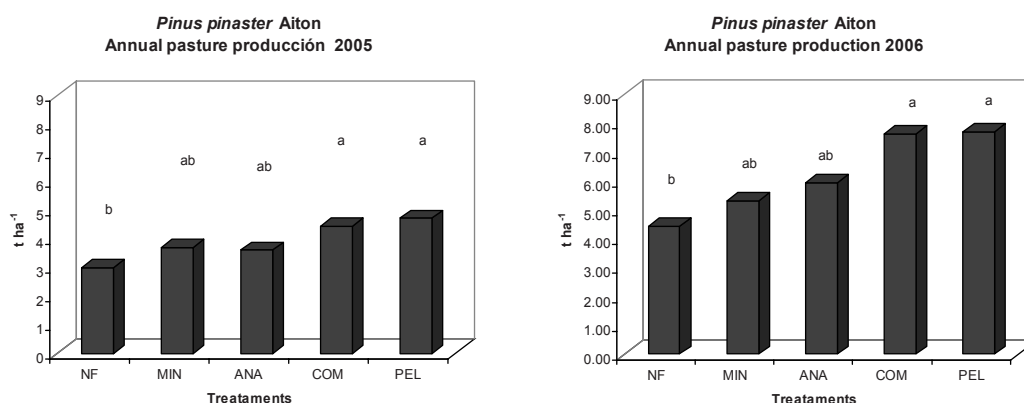


Figure 2. Annual pasture production under of years 2005 and 2006 for the different treatments (NF: no fertilization, MIN: mineral, ANA: anaerobic, COM: compost, PEL: pelletized).

## Conclusions

Pasture and tree growth depends on previous land use and soil type, which modifies the response of both pasture and trees to fertilization treatments. Composted and pelletized sewage sludge were shown to promote tree growth and pasture production on abandoned land.

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## Changes in some morphological characteristics of reed canary-grass (*Phalaroides arundinacea* L.) during primary growth

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### Abstract

This paper presents measurements of some morphological characteristics of *Phalaroides arundinacea* L. during primary growth, based on 4 years (2004-2007) of field investigations. The extended shoot height (ESH), the number of leaves (NL) per shoot, and number of dead leaves (NDL) per shoot, amounted to 89.06 cm, 8.80 leaves and 3.33 leaves, respectively. There were 5.33 active or live leaves per shoot on average during the investigation period. Sum of active leaf lamina length (SALL), the index of leafiness (IL) and ratio of SALL to ESH were increasing to the middle of investigation period then started to decrease until early June. Between-years differences were recorded, with a significantly weaker vegetative growth (NL, NAL and SALL) in 2007 which could be related to a shortage of water in that year.

**Keywords:** reed canary-grass, morphological characteristics, primary growth

### Introduction

Harvestable grass yield during the defoliation of primary growth in spring is constituted basically from the stems (vegetative or generative) and leaves (sheath and lamina). In the case of late utilization these are supplemented with inflorescence. Spring development of these grass parts on a tiller shows a well known pattern (Robson *et al.*, 1989; Nelson and Moser, 1995). After a tiller has been differentiated and starts to grow, the vegetative stems remain short (consisting of nodes and very short internodes) hidden under the leaves' sheaths and new leaves develop consistently from the nodes within certain periods of time. The leaves which then develop have a limited life span (e.g. 3 weeks) after which they die and are replaced by the new leaves. This procedure is terminated by generative development, whereby the vegetative shoot elongates, overgrows the last leaf sheath, and finally differentiates into the reproductive structure, the inflorescence. To quantify some morphological characteristics of reed canary-grass (*Phalaroides arundinacea* L.) during primary growth, a research programme was conducted at Debrecen University, Hungary.

### Materials and methods

Morphological measurements of reed canary-grass were made under field conditions at the demonstration garden of Debrecen University, Agricultural Centre. The ecological conditions of the site are as follows: soil structure is high fertility loam (chernozem); average climatic conditions (last forty years) to the middle of June – precipitation 227.1 mm, T-sum of mean daily temperatures 1,244.5 °C, sum of daily sunshine hours day 889.2 hours.

Individual shoots of reed canary grass (n = 30) were randomly selected from a pure stand of the cultivar 'Keszthelyi 52' and tagged with a plastic ribbon. Leaves on the shoot were numbered upwards from ground level; the plastic ribbon tags were placed between the last dead leaf and the first live leaf on the first day of measurement. The measurements were then repeated seven times (altogether 8 measurements per year) at weekly intervals during the primary growth between mid-April and early June.

Primary data recorded on each shoot and on each occasion were:

- extended shoot height/length (ESH),
- number of dead leaves (NDL) (a leaf was considered dead if more than half of the leaf lamina from the tip was withered),
- lamina length (LL) of the live leaves (cm).

During the evaluation procedure, secondary data were generated from the primary data:

- number of leaves (NL) per shoot developed to date (pieces),
- number of dead leaves (NDL) per shoot on each date (pieces),
- number of active leaves (NAL) per shoot on each date (pieces),
- sum of active/live leaf lamina (SALL) lengths on the shoot (cm) on each date,
- index of leafiness (IL) ratio of SALL to ESH, which is used as an indicator for the leafiness of a grass.

Data obtained were statistically analysed using General Linear Model of the SPSS 13.0 for Windows (2004) software.

## Results and discussion

The extended shoot height (ESH), the numbers of leaves per shoot, and dead leaves per shoot were increasing through the investigation period and amounted to 89.06 cm, 8.80 pieces and 3.33 pieces, respectively (Table 1). On average, there were 5.33 active/live leaves per shoot during the investigation period of primary growth. The sum of active leaf lamina length and the index of leafiness were increasing in the beginning, and amounted to 145.0 cm and 2.26, respectively. Later on, they were decreasing to 126.8 cm and 1.65, respectively. A very similar pattern was found for the average size (cm) of the live leaf lamina length. In the chronology of emergence of leaves the lamina was 8.5 ( $s = 3.9$ ), 15.3 ( $s = 6.4$ ), 21.7 ( $s = 6.9$ ), 25.6 ( $s = 7.0$ ), 27.0 ( $s = 7.3$ ), 26.3 ( $s = 7.3$ ), 22.8 ( $s = 8.0$ ), 19.0 ( $s = 9.5$ ), 17.8 ( $s = 8.4$ ), 16.1 ( $s = 8.7$ ), 16.3 ( $s = 7.7$ ) and 12.0 ( $s = 4.2$ ) for the leaves 1st, 2nd, 3rd, 4th, 5th, 6th, 7th, 8th, 9th, 10th, 11th and 12th on the shoots from the ground, respectively. Data show that the successive leaves in the beginning of primary growth period were bigger and bigger until leaf size culminated. Later on, successive leaves decreased in size. The last leaf (12th) was only 44% the size of the biggest leaf (5th one).

Table 1. Morphological characteristics of reed canary-grass (2004-2007) during primary growth. ESH = extended shoot height, NL = number of leaves per shoot, NDL = number of dead leaves per shoot, NAL = number of active leaves per shoot, SALL = sum of active/live leaf lamina lengths on the shoot, IL = index of leafiness.

Dates	15 Apr	22 Apr	29 Apr	6 May	13 May	20 May	27 May	3 June	Mean	LSD <sub>5%</sub>
ESH, cm	48.78	55.16	60.82	68.53	74.27	79.76	84.69	89.06	71.06*	4.13
NL per shoot	5.56	6.27	6.85	7.41	8.07	8.50	8.67	8.80	6.99*	0.43
NDL per shoot	0.34	0.59	1.05	1.45	1.90	2.39	2.93	3.33	1.66*	0.31
NAL per shoot	5.22	5.68	5.81	5.96	6.17	6.10	5.74	5.46	5.33*	0.38
SALL on a shoot	95.09	113.60	127.78	139.20	145.03	143.92	134.99	126.89	128.31*	10.51
IL	2.05	2.16	2.26	2.16	2.12	2.02	1.78	1.65	1.99*	0.18

\* $P < 0.001$ .

Extended shoot height was outstandingly high (114.9 cm) in 2006 and remarkably low (49.24 cm) in 2007 (Table 2). Number of leaves developed, number of active leaves and SALL per shoot were lower in 2007.

Table 2. The differences in the measured morphological characteristics of reed canary-grass among the experimental years. ESH = extended shoot height, NL = number of leaves per shoot, NDL = number of dead leaves per shoot, NAL = number of active leaves per shoot, SALL = sum of active/live leaf lamina lengths on the shoot, IL = index of leafiness.

Morphological characteristics	2004	2005	2006	2007	Mean	LSD <sub>5%</sub>
ESH, cm	58.58	57.82	114.90	49.24	71.06*	8.26
NL per shoot	8.15	7.61	7.78	6.52	6.99*	0.85
NDL per shoot	1.67	1.49	1.89	1.95	1.66*	0.61
NAL per shoot	6.48	6.12	5.89	4.57	5.33*	0.76
SALL on a shoot	144.74	132.61	144.15	91.75	128.31*	21.02
IL	2.54	2.30	1.35	1.92	1.99*	0.37

\* $P < 0.001$ .

Annual precipitation showed the greatest differences between years (Table 3). For two years (2004 and 2005), it was above the last 40-years average. In 2006, the experimental field received twice as much rainfall as the average. In 2007 there was a huge shortage of precipitation in spring time. Both temperature conditions and accumulated sunshine hours were more balanced than rainfall. However, the sum of sunshine hours in 2006 was remarkably lower. Both sum of temperature and sum of sunshine hours were remarkably higher in 2007.

Table 3. The relative availability (%) of key climatic conditions (last 40 year average = 100%) for grass growth in the experimental years.

Years	Rainfall <sup>a</sup> , mm	Temperature sum <sup>a</sup> , °C	Sum of Sunshine hours <sup>a</sup> , h	Nature of weather in the year cf. average
2004	105 <sup>b</sup> -151 <sup>c</sup>	104-115	97-102	rainy, slightly warm, average sunshine
2005	96-130	99-108	107-115	slightly rainy and sunny, average temperature
2006	187-221	107-112	88-92	extremely rainy, slightly warm, cloudy
2007	74-94	151-213	119-128	reasonably dry, extremely warm, very sunny

<sup>a</sup>from 1st of January; lowest minimum <sup>b</sup>and highest maximum <sup>c</sup>values in the experimental periods.

These differences in the weather conditions can explain the differences among the years in the measured characteristics of reed canary-grass. The significantly higher and lower ESH for 2006 and 2007, respectively, may be attributed to the surplus and shortage of rainfall in these years. It seems that the relatively lower sunny hours in 2006 did not depress the spring growth of reed canary grass in that year. The significantly weaker vegetative growth (NL, NAL and SALL) in 2007 might definitely have been controlled by a shortage of water.

## Acknowledgements

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# Winter survival, yield performance and forage quality of *Festulolium* cvs. for Norwegian farming

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## Abstract

*Festulolium* cvs. have been tested throughout Norway. If high forage quality is required, Hykor and Fojtan (*L.m. x F.a.*) should be cut frequently (at least 3 cuts year<sup>-1</sup>) due to an early heading in the first cut and a rapid reduction in forage energy concentration in the leafy regrowth. Hykor is more persistent and high yielding than Fojtan. In mixtures *Festulolium* cvs. make a more stable crop and are more flexible in use than as pure stand. Felopa and Perun (*L.m. x F.p.*) are less winter hardy than Hykor and they are early heading cvs. In regions without a harsh winter climate these cvs. are competitive to perennial ryegrass. Forage quality is high. New candidate cultivars are promising but still at an early testing stage.

Keywords: *Festulolium*, winter survival, yield, forage quality, plant development

## Introduction

*Festulolium* is defined as hybrids resulting from the crossing of a species of the genus *Festuca* with a species of the genus *Lolium* (*Festuca* spp.  $\times$  *Lolium* spp.). In the OECD List of Varieties it is described as '*x Festulolium*' and the names of the species used in the hybrid should be mentioned. There is interest in *Festulolium* cultivars (cvs.) in Norway mostly because of high yield and reliable winter survival of cultivars from East European countries. Graminor AS (Ltd), the Norwegian plant breeding company, has *Festulolium* as one of their breeding priorities and their main objective is to transfer winter hardiness from meadow fescue (*F. pratensis*) into ryegrass (*Lolium* spp.). A few candidate cultivars are being tested in field, however, most of the breeding material is still in an early breeding stage. To get knowledge of survival ability, yield levels and forage quality of different cultivars for recommendations, research projects are currently being undertaken in Norway to assess *Festulolium* cvs. in pure stands or in mixtures with timothy (*Phleum pratense*), meadow fescue, perennial ryegrass (*L. perenne*) and red clover (*Trifolium pratense*) as well as in trials to explore the potentials of *Festulolium* cvs. in grazing and silage harvesting regimes. An additional project started in 2006, including foreign cvs. and Norwegian candidate cvs., will give new knowledge on plant development and leaf : stem ratios through the growth season.

## Materials and methods

Three experimental series with field trials conducted in Norway are presented. (1) Two contrasting *Festulolium* cvs., Hykor (*L. multiflorum x F. arundinacia*) and Felopa (*L.m. x F.p.*) compared to a commonly used seed mixture containing 50% timothy, 30% meadow fescue, 10% perennial ryegrass and 10% red clover, were established in field trials in 2003 (7 locations) and 2004 (8 loc.). (2) Cultivars of *Festulolium*, *Lolium* hybrid (*L. x boucheanum*) and perennial ryegrass were established in 2005 at 14 locations to explore the potentials of yield, forage quality and winter survival when exposed to two harvesting regimes of either three or five cuts per season. Growth start in 2006 was estimated according to Bonesmo (2004) and day degrees during the growth season 2006 calculated using inputs of daily measurements of temperature from automatic weather stations representative of growth

conditions for the established field trials. For series 1 and 2 all cultivars were cut at the same date in each trial. For all series plant material was dried at 60 °C for 48h (dw) and quality parameters analysed with NIRS. (3) A field trial with *Festulolium* cvs. (Hykor, Felopa) and candidate cvs. was established in 2006 at Fureneset, West Norway (61.29°N, 5.04°E) with a four-cut system with individual cutting at early heading in 2007. Samples of at least 45 shoots per plot (3 replicates) were sorted into leaves and stems at each harvest, dried and weighed.

## Results and discussion

Winter survival as spring ground cover for cvs. or mixture (Table 1) demonstrated the good survival ability of Hykor, which in both years of series 1 (2004-06 and 2005-07) was better than the mixture of mostly timothy and meadow fescue, species that are normally very winter hardy in Norwegian agriculture. Felopa showed much lower and less stable survival compared to Hykor. Similar differences between Hykor and Felopa was shown in the spring 2006 (Table 1, series 2), following a very hard winter with heavy and long snowfall in some regions or with frost in late winter following a mild mid-winter in other regions. The differences between Hykor and the other *Festulolium* cvs. were less clearly expressed here.

Table 1. Winter survival and DM yield as mean values of three harvest years (2004-06, 2005-07) and in 1<sup>st</sup> year of ley (2006) for cvs. of *Festulolium* (F), *L. x boucheanum* and *L. perenne*.

Series		1		2	1		2	
Years of exp. (harvest year)		2004-06	2005-07	2006(1)	2004-06	2005-07	2006(1)	2006(1)
No. of field trials (cuts/year)		7	7	14	7 (3)	8 (3)	14 (3)	14 (5)
Cvs / mixture	Species	Spring ground cover (%)			Kg DM ha <sup>-1</sup>			
Seed mixture		73	65	-	11,622	10,481	-	-
Hykor	F, <i>L.m. x F.a.</i>	78	76	68	12,325	11,794	10,118	8,345
Felopa	F, <i>L.m. x F.p.</i>	49	40	55	10,868	9,510	10,248	8,075
Fojtan	F, <i>L.m. x F.a.</i>	-	-	60	-	-	9,246	7,557
Perun	F, <i>L.m. x F.p.</i>	-	-	66	-	-	11,178	8,567
FuRs9806	F, <i>L.p. x F.p.</i>	-	-	73	-	-	10,428	8,424
Fenre	<i>L. x boucheanum</i>	-	-	68	-	-	10,892	8,618
Napoleon	<i>L. perenne 4x</i>	-	-	74	-	-	10,634	8,705
P value		<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05
LSD 5%		6.77	5.05	5.5	453	368	593	495

Mean total dry matter (DM) yield over three harvest year (Table 1, series 1) showed that Hykor performed better than both the seed mixture and Felopa, partly due to better ground cover and an early heading time compared to the other cultivars. The 2005-07 field trials of series 1 were located in regions with harder winter conditions than the 2004-06 trials, which may explain the lower level of DM yield. In the first harvest year of series 2, the 5-cut system produced 80% of the DM obtained in the 3-cut system (Table 1). Hykor showed lower DM yield than the other cvs. except for Fojtan in the 3-cut system, and for Fojtan and Felopa with 5 cuts; however, significantly only for Perun and Fenre at 3 cuts. The yield level in 2006 may express the yield potentials of the cvs. in regions where winter survival is not the crucial factor. Perun was high yielding in both management systems. Fojtan, with the similar parental combination as Hykor and specifically bred for grazing purposes, showed neither survival ability nor yield potential to be recommended along with the other *Festulolium* cvs. tested. Forage energy concentration (FEm kg<sup>-1</sup> DM) for all cuts in 3- and 5-cut system of series 2 (Table 2) showed the necessity of early harvest of *Festulolium* cvs. Significant negative correlations between day degrees and forage energy concentration (data not shown) were found for the three silage harvests and for the first three and the last cut in the simulated

grazing regime. This is particularly important for Hykor due to the early heading, and Hykor mixed with several species produced a more flexible crop with respect either to yield or forage quality (Østrem and Hamar 2005).

Table 2. Forage energy concentration (FEm kg<sup>-1</sup> DM) in 1st year of ley, in 3 cuts of silage harvests and 5 cuts of simulated grazing system. Mean of 14 field trials.

Management	Normal			Simulated grazing				
Cut	1	2	3	1	2	3	4	5
Hykor	0.88	0.85	0.88	0.98	0.93	0.90	0.90	0.92
Fojtan	0.87	0.84	0.87	0.98	0.92	0.90	0.90	0.92
Perun	0.90	0.81	0.90	1.03	0.95	0.90	0.92	0.95
Felopa	0.91	0.82	0.92	1.04	0.94	0.92	0.92	0.96
FuRs9806	0.94	0.85	0.92	1.02	0.93	0.91	0.91	0.95
Fenre	0.91	0.83	0.91	1.03	0.93	0.90	0.91	0.94
Napoleon	0.92	0.83	0.90	1.01	0.94	0.90	0.91	0.94
P value	<0.05	<0.05	<0.05	<0.05	0.05	0.41	<0.05	<0.05
LSD 5%	0.024	0.020	0.015	0.025	0.016	0.018	0.016	0.015

Differences in yield and especially in forage energy concentration may be explained by differences in heading time and plant development during the growth season. A harvest by cultivar interaction was found for leaf vs. stem proportions during the 2007 growth season in series 3. Hykor differed from all other tested cultivars by high leaf:stem ratio increasing from 2.1 (cut 1) to 4.1 (cut 4). Fure meadow fescue displayed a similar development as Hykor, though with more stems, especially in cut 1. In contrast, Felopa displayed a low leaf:stem ratio of 1.1, 0.6 and 0.8, respectively, for the first three harvests. Among the candidate cvs. FuRs0136 had high stem production during all four harvests (1.0 as mean for four cuts) due to the Italian ryegrass in its parental combination. Hykor and Fojtan have a very similar growth development with early heading and by only producing leaves in the regrowth. Hykor rarely made more than two leaves per shoot (series 3, data not shown) which made the cultivar very different from all other *Festulolium* cvs. Also in the regrowth (Table 2) the forage energy concentration was generally lower for Hykor and Fojtan compared to cultivars containing more stems in the total plant biomass. The high DM yield of these cvs. combined with few leaves per shoot imply that the leafy biomass are old and may thus have reduced quality.

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# Recovery of fertilizer N in grassland communities of different diversity and composition

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## Abstract

A  $^{15}\text{N}$ -fertilization study was made in an experiment where plant diversity was varied between one and twelve species. Fertilizer N ( $50 \text{ kg N ha}^{-1}$  as  $\text{NH}_4\text{NO}_3$ ) was added to the plant communities in early June of two consecutive years. On average 70% of fertilizer N was recovered in the above- and below-ground plant fractions over the two years. Recovery of fertilizer N in plant biomass was correlated to grass biomass as proportion of total biomass. However, the difference in  $^{15}\text{N}$  recovery was much smaller than the difference in total biomass N between plant communities. It is likely that N losses due to denitrification or leaching were small, due to the dry weather during the two growing seasons and frozen ground in winter.

Keywords: Biodiversity, nitrogen,  $^{15}\text{N}$  recovery in plants

## Introduction

Efficient use of fertilizer N is necessary in sustainable agriculture. From the farmers' point of view, N losses are both wasteful and cause acidification of the agricultural soil. In other ecosystems, N losses cause environmental hazards such as eutrophication of waters by nitrate leaching, and  $\text{N}_2\text{O}$  emissions contribute to the greenhouse effect and stratospheric ozone depletion. This study aims to identify plant community traits that influence fertilizer N utilization by using a well established field experiment where species richness and plant functional group composition was varied.

## Materials and methods

The field experiment was established as a part of the European BIODEPTH-project (Biodiversity and Ecological Processes in Terrestrial and Herbaceous Ecosystems) in Umeå, Sweden ( $63^\circ 49'\text{N}$ ,  $20^\circ 17'\text{E}$ ). Plant communities (72 plots,  $2 \times 5 \text{ m}$ ) with 1, 2, 4, 8 or 12 species were established by sowing on agricultural land in 1996. There were four grasses: *Dactylis glomerata* L. cv. Laika, *Festuca ovina* L., *Phalaris arundinacea* L. cv. RF9402 and *Phleum pratense* L. cv. Jonatan; four legumes: *Lotus corniculatus* L., *Trifolium hybridum* L. cv. Stena, *Trifolium pratense* L. cv. Betty and *Trifolium repens* L. cv. Undrom, and four non-leguminous forbs: *Achillea millefolium* L., *Leucanthemum vulgare* (L.) Lam., *Ranunculus acris* L. and *Rumex acetosa* L. There were also two bare plots. The species composition of the plots was maintained by regular weeding during the summers, except for four unweeded and unsown control plots where plant communities were established spontaneously. Resowing of sown plots was done in spring when necessary. Further details are found in Palmborg *et al.* (2005). The experiment had not been fertilized from 1996, but in early June 2002 and 2003 one square meter subplots, limited by plastic boards reaching 35 cm down into the soil, in 49 of the plots were fertilized with  $^{15}\text{NH}_4^{15}\text{NO}_3$  (5 atom %  $^{15}\text{N}$  excess) corresponding to  $5 \text{ g N m}^{-2}$  each year. The fertilizer was applied with a watering-can, dissolved in the equivalence of 2 mm precipitation of water, followed by application of 2 mm of pure water to wash down the solution from the vegetation. The subplots were situated within a  $2 \times 2.2 \text{ m}$  area fertilized

with the same rate of unlabelled  $\text{NH}_4\text{NO}_3$ . These plots represented the full variation in species richness and in the functional groups: grasses, legumes and non-leguminous forbs.

Each year, in mid-August, plant biomass above 5 cm was sampled from an area of 50 x 20 cm and sorted to species. In 2003, a 20 x 20 cm sample of the remaining aboveground biomass and litter was taken. The same year, after cutting of the entire plots to 5 cm, four soil samples were taken from 0-20 cm ( $\text{Ø} = 29$  mm), and 20-40 cm ( $\text{Ø} = 21$  mm) depth in each plot and pooled. Roots were extracted from the soil by washing on a 0.5 mm mesh and flotation in water to remove sand. All plants that were removed by weeding of the plots were collected and pooled by plot and year. All plant material was dried at 60 °C and ground in a ball mill before analysis of  $^{15}\text{N}$  abundance and N concentration. Percent  $^{15}\text{N}$  recovered was calculated as in Hauck and Bremner (1976) for the four plant fractions; biomass above 5 cm with weeds excluded (harvest), 0-5 cm (stubble), roots 0-40 cm (roots), and weeds removed by weeding plus weeds in the harvest (weeds).

Data was analysed by ANOVA and Fishers test of least significant difference and the general linear model module in SYSTAT 11. The significance level used was  $P < 0.05$ .

## Results and discussion

Above-ground biomass was often higher in multi-species communities than in any of the monocultures of the species being part of the community. The total plant N was positively correlated to the number of sown species in the plot and to legume biomass and grass biomass as proportions of total biomass. Total recovery of fertilizer N was generally high (Table 1) and there were small differences in total recovery of fertilizer N among the plant communities. Among sown plots the only trait that was significantly correlated to total recovery of fertilizer N was proportion of grass in the harvest (adj  $R^2 = 0.15$ ,  $P = 0.006$ ). The high total recovery was probably due to the dry weather during the two growing seasons. Precipitation in the growing season of 2002 was 57% of normal and before harvest 2003 70% of normal. In Finland there was higher  $^{15}\text{N}$  recovery in *Phalaris arundinacea* after a dry early summer than after a wet summer (Partala *et al.*, 2001). Another reason could be that the  $^{15}\text{N}$  fertilization was repeated in 2002 and 2003 in our experiment. Thus, some of the  $^{15}\text{N}$  recovered in 2003 could have been mineralized from litter from the year before. A higher mean recovery of fertilizer N in the 2003 harvest (38%) than in the 2002 harvest (32%) indicated that this was the case.

Table 1. Mean recovery of fertilizer N in different biomass fractions and total biomass and total biomass N. For harvest and weeds, data from two years was summed. Means within a column with the same letter are not significantly different.

Plots (n)	Harvest (% recovery)	Stubble (% recovery)	Roots (% recovery)	Weeds (% recovery)	Total biomass (% recovery)	Total biomass N content (g N m <sup>-2</sup> )
Bare (2)		1 a	3 a	40 c	44 a	16 a
Grasses (6)	26 ab	16 b	25 bc	14 b	81 c	30 a
Legumes (8)	31 bc	17 b	17 b	6 a	71 b	41 b
Forbs (6)	18 a	18 ab	21 bc	6 a	63 ab	20 a
Mixed (23)	36 bc	9 c	24 c	7 a	74 b	40 b
Unweeded (4)	43 c	9 abc	25 bc		77 b	36 b

When plant communities were divided into the groups in Table 1, there were significant differences between groups for all biomass fractions. Recovery of fertilizer N in the harvest was largest in mixed plots and unweeded plots. Multiple regression using only sown plots revealed that species richness, proportion of legumes in the harvest and proportion of grass in the harvest were positively and significantly correlated to recovery of fertilizer N in the

harvest. However, species richness was significant only if weeds were not counted as part of the harvest. The aboveground recovery was comparable to studies of grass swards, where fertilizer N recovery ranged from 35% to 68% in the aboveground biomass of grasses (Lyngstad 1991; Partala *et al.*, 2001; Williams *et al.*, 2001).

Forbs had a smaller total fertilizer recovery than grasses, and a comparably larger part of the recovery was in the stubble in forbs. The flowering shoots of some of the forbs were already senescent at the time of the harvest, and thus they probably had passed their maximum content of N in the biomass. A large part of the leaves were close to the ground and thus collected in the stubble fraction. Recovery of fertilizer N in roots was generally high and it was larger in mixed plots than in legume plots. In other studies 2-19% of fertilizer N was recovered in roots and rhizomes of *Phalaris arundinacea* (Partala *et al.*, 2001) and 5-22% in roots of seed crop grasses in New Zealand (Williams *et al.*, 2001). The high recovery in roots in our study could be due to the large mass of roots that had accumulated during six years without fertilization prior to the fertilization treatment.

The bare plots had the smallest total recovery of N fertilizer, although it was surprisingly high. Bare plots were frequently weeded and since weeds were removed when small they had a high N concentration and a high  $^{15}\text{N}$  atom %.

## Conclusions

Regardless of plant functional group composition, N-losses were low under the conditions of this study. However, a larger part of the fertilizer N was allocated to the harvest of sown species in species-rich communities where legumes or grasses were dominating, than in communities rich in forbs. Our study also shows that weeds removed in diversity experiments can contain a considerable part of the plant available N and should, therefore, not be neglected in nutrient budgets.

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# Nitrogen efficiency of farm manure on permanent grassland in mountainous regions

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## Abstract

By means of exact field trials comprehensive studies on the nutrient efficiency of different farm manure systems have been carried out on permanent grassland at three different sites in Austria. Slurry, solid manure and liquid slurry from cattle were applied on three-cut and four-cut meadows in different doses for a period of six years. Analyses on yield, forage quality and floristic diversity were carried, as well as investigations on nutrient fluxes and nutrient efficiency. Compared with mineral fertilizer systems, farm manure showed a high efficiency, which is not only based on the content of main nutrients but also on some additional effects. The results clearly indicate the high efficiency of farm manure on permanent grassland and show that sufficient yields and forage quality can be reached without the use of any additional mineral nitrogen.

Keywords: farm manure, N-efficiency, nitrate directive, permanent grassland

## Introduction

Due to the regulations of the European Nitrate Directive, the yearly application rate of manure on grassland is limited by the load of nitrogen in order to avoid negative impacts on the environment. The amount of 170 kg N ha<sup>-1</sup> ex storage represents the general limit, which in Austria can be extended to 230 kg in compliance with some special obligations (Aktionsprogramm, 2008). Most of the Austrian grassland and dairy farmers are included in the Austrian agri-environmental programme ÖPUL. Therefore, the sustainable and efficient use of farm manure is of great importance and, together with the optimal use of home-grown forage, is an important strategy of low input farming. Due to unavoidable N-losses, mostly occurring via NH<sub>3</sub>-volatilization, the nitrogen efficiency of farm manure is lower than that of mineral nitrogen fertilizer. Following the official Austrian guidelines for an appropriate fertilization (BMLFUW, 2006), the assumed nitrogen efficiency of farm manure also considers unavoidable N-losses for stable, storage and application, and is additionally reduced due to a low yearly efficiency. By means of comprehensive field experiments on three Austrian grassland sites, basic aspects of nitrogen and system efficiency of farm manure have been considered and critically discussed.

## Material and methods

Different fertilizer systems have been tested on permanent grassland under a medium intensive three-cut and a highly intensive four-cut regime, each on three Austrian sites. The farm manure was derived from dairy cattle and was applied at two different intensity levels and split in three or four doses, respectively, depending on the cutting frequency. The applied nitrogen amount is based on an ex-storage level, which considers unavoidable N-losses for stable and storage according to the European Nitrate Directive (EU-Nitratrictlinie, 1991). Each of the variants in the high intensity system received an additional amount of mineral nitrogen at the rate of 50 kg ha<sup>-1</sup> year<sup>-1</sup>.

Table 1. Description of the experimental sites.

Site	Altitude in m ASL	Ø yearly temperature	Ø yearly precipitation
Kobenz	627	8.2 °C	856 mm
Winklhof	490	8.2 °C	1400 mm
Gumpenstein	710	6.8 °C	1010 mm

Table 2. Treatments and average nutrient application rates in the farm manure experiment during the project period (2001-2006).

Intensity systems/ Treatments	number of cuts year <sup>-1</sup>	Ø nutrient application (kg ha <sup>-1</sup> year <sup>-1</sup> )		
		N <sub>ex storage</sub>	P	K
NPK	3	92.2	20.2	91.4
Slurry	3	94.5	13.6	84.0
Stable manure + liquid slurry	3	103.8	28.5	176.3
Composted manure + liquid slurry	3	118.8	31.6	185.8
NPK	4	234.3	40.3	182.9
Slurry + NPK	4	236.6	26.1	162.4
Stable manure + liquid slurry + NPK	4	239.0	49.4	317.9
Composted manure + liquid slurry + NPK	4	256.5	54.1	310.9

The field experiments were established in the year 2000 and recordings and analyses were carried out from 2001 to 2006 for all variants, each with four replications. A strong focus was given to the nitrogen efficiency of farm manure, which was related to yield production by means of the following equation:

$$N_{\text{eff-yield}} \text{ (kg DM kg N}^{-1}\text{)} = \frac{\text{yield of variant (kg DM ha}^{-1}\text{ year}^{-1}\text{)}}{\text{kg N-Input of variant (kg N ha}^{-1}\text{ year}^{-1}\text{)}}$$

The results of this calculation were placed in relationship to the N-efficiency of the particular mineral nitrogen variant within the two intensity systems, which were fixed at 100% each.

## Results and discussion

In both of the intensity systems and at all sites a high productivity level could be observed at the beginning of the project period followed by a strong decline, which was mainly caused by drought in 2002 and 2003 (Figure 1).

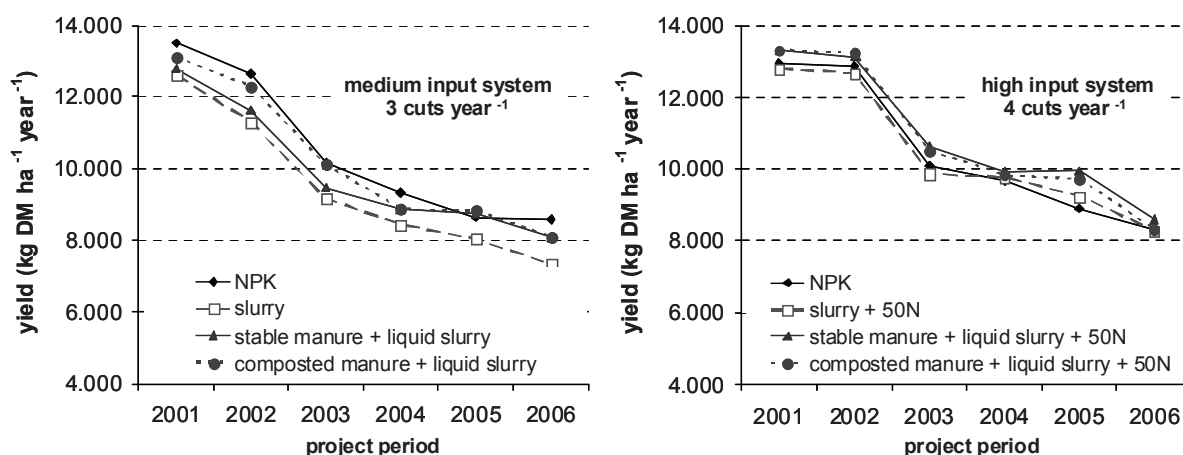


Figure 1. Yield productivity (kg DM ha<sup>-1</sup> year<sup>-1</sup>) of different variants during the project period (average of all three sites)

All variants were similarly influenced but at a different yield level, which ranged between 9,480 and 10,920 kg DM ha<sup>-1</sup> year<sup>-1</sup> for the total observed period. It is evident that there was



just a minor difference between the yield level of the medium and the highly intensive systems even though the nutrient input varied strongly. Within the two intensity systems significant differences were found between the treatments. In the 3-cut system the mineral NPK variant showed the highest average yield during the project period, whereas in the intensive 4-cut system the variant receiving stable manure + liquid slurry + mineral NPK performed best.

Table 3. Relative nitrogen efficiency (%) of different fertilizer systems on three sites (average of 2001-2006).

Intensity systems/ Treatments	Kobenz	Winklhof	Gumpenstein	Assumed efficiency
Mineral NPK	100	100	100	100
Slurry	84	90	92	61
Stable manure + liquid slurry	75	87	89	38
Composted manure + liquid slurry	73	79	86	21
Mineral NPK	100	100	100	100
Slurry + mineral NPK	96	101	99	61
Stable manure + liquid slurry + mineral NPK	100	102	103	38
Composted manure + liquid slurry + mineral NPK	91	96	97	21

The observed N-efficiency of farm manure ranges between 73% and 92% compared with mineral nitrogen (= 100%) in the medium intensive 3-cut system. A strong variation occurred both within the project period of 6 years and between the tested sites. The observed N-efficiency was significantly higher than the assumed N-efficiency according to the Austrian guidelines for an appropriated fertilization (BMLFUW, 2006). This result was also confirmed in the high intensity 4-cut system with an even higher relative N-efficiency of the used farm manure (91% to 103%). Within this group the stable manure system resulted in the highest efficiency at all three sites. The absolute N-efficiency was significantly higher in the medium intensive system (101 kg DM kg N<sup>-1</sup>) compared with the high intensive system (42.5 kg DM kg N<sup>-1</sup>). This clearly shows that high nutrient inputs do not automatically result in high yields. Therefore, the additional use of mineral nitrogen on grassland especially has to be seriously reconsidered.

## Conclusions

The current procedure for the valuation of farm manure which is regularly used on permanent grassland should be reconsidered. It is evident that the efficiency of farm manure, which is not only based on the content of nitrogen but also on other nutrients and on additional effects (e.g. organic matter input), is higher than assumed. This is especially so in the case of stable manure and composted manure, whose efficiency is strongly underestimated. This approach reduces both the real and the ideological value of farm manure, which should be the main nutrient source in sustainably managed grassland farms.

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# Legume-cruciferae mixture as a multi-purpose crop in almond orchards in a semiarid climate

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## Abstract

Cover crops are known to play an important role in soil protection and weed control, but they can also serve as a food resource for livestock in semiarid climates. We studied the performance of a mixture composed of *Medicago sativa* L., *Vicia ervilia* (L.) Willd. and *Moricandia arvensis* (L.) DC. in a two-year crop-fallow rotation in an almond orchard, under different grazing regimes. Biomass production and proportion by weight of weeds and of each species were measured during two periods: crop (2006) and fallow (2007). Grazing periods were spring and/or summer during the crop year. The results showed that biomass production was 30% greater for the fallow than for the crop period. Spring grazing in 2006 did not diminish the 2007 biomass production. Weeds constituted more than 54% of the total dry weight for the crop but around 27% for fallow. The selected species were resistant to grazing and showed a high self-reseeding ability, especially, *V. ervilia*. We conclude that this legume-cruciferae mixture constitutes an important source for livestock feeding that can be consumed in spring and/or summer; it may maintain the forage production for several years, and it can be successful in weed control.

Keywords: cover crop, fodder, biomass, weed control, self-reseeding

## Introduction

In south-eastern Spain, almonds, cereal and sheep are the main components of marginal dry-farming systems. In this area, almond orchards can be very sensitive to soil erosion (van Wesemael *et al.*, 2003). Nevertheless, intensive tillage is practised by farmers to increase water infiltration and reduce weed competition. Moreover, the low profitability of the almond crop in these systems is leading to land abandonment. Therefore, new sustainable alternatives to these orchards are needed.

Cover crops have been demonstrated to be efficient for increasing soil organic matter, improving soil structure, and reducing surface runoff in comparison to long-term cultivation (Haynes, 1980) and, consequently, in the control of soil erosion. Additionally, cover crops may further serve multiple functions such as weed control, refuge for beneficial arthropods, or forage for livestock. The latter may be one of the most profitable functions for the above-mentioned marginal areas, due to the increase of forage availability. Legumes are appropriate for this multi-purpose as they provide organic matter and N to the soil and they have a high nutritive value for livestock feeding.

We studied the performance of a mixture composed of two legumes and one cruciferae: *Medicago sativa* L., *Vicia ervilia* (L.) Willd. and *Moricandia arvensis* (L.) DC., in a two-year crop-fallow rotation in an almond orchard, under different grazing regimes.

## Materials and Methods

The study was conducted in Huéscar (northern Granada province, SE Spain, UTM coordinates: 305-535799.04-4192415.19, and 990 m a.s.l.). Trials were performed in a petric calcisol soil (FAO, 2006), with a slope of approximately 2%.

The climate in this area is continental semiarid Mediterranean, with mean yearly temperature and precipitation of 13 °C and 481 mm, respectively.

The experiment was carried out over two growing seasons (2006 and 2007) in a 30-year-old almond orchard (*Prunus dulcis* (Miller) cv. 'Verdiere') where tree spacing was 10 m x 14 m. Prior to the establishment of the plots, the entire experimental area was ploughed and disked to create a suitable seedbed. In January 2006 six plots (40 x 42 m) were established. All of them were sown by hand with a seed mixture composed of *Medicago sativa* L. cv. 'Aragón' (24 kg ha<sup>-1</sup>, 12 million seeds), *Vicia ervilia* (L.) Willd. (57 kg ha<sup>-1</sup>, 1.14 million seeds) and *Moricandia arvensis* (L.) DC. (0.7 kg ha<sup>-1</sup>, 1.4 million seeds). Seeds were covered using a heavy bar attached to a tractor. Three randomly selected plots were fenced to allow controlled grazing and, in May 2006, 382 sheep grazed for a total of 2 h per plot. In July, fences were removed and sheep were allowed to graze the whole area 2 h every 7 or 10 days, approximately, until September. The experimental area was left fallow, without livestock, until July 2007.

Biomass production of the cover crop was estimated by hand-clipping 6 randomly selected 0.25 m<sup>2</sup> quadrats in each plot the day before the grazing period in May 2006 and in May 2007. Three homogenized sub-samples of biomass from each plot were oven dried at 60 °C to constant weight to determine dry weight. Averaged data were calculated on a per-hectare basis. Three of the clipped forage samples from each plot were used to determine weed biomass and species composition, after hand separation.

Statistical analysis of biomass production and of the dry weight percentage of each species was performed with a two-way ANOVA. Differences among treatments were detected by the Fisher-LSD test.

## Results and discussion

Fodder yield was greater in the fallow year (2007) than in the crop year (2006) (Table 1), probably due to higher precipitation during 2007, especially in spring. Moreover, spring grazing during 2006 did not affect fodder yield in 2007 (grazing vs. no grazing, Table 1).

*Vicia ervilia* showed the highest dry-weight percentage in 2006 (Table 1), probably due to its better adaptation to the pedoclimatic conditions. *Medicago sativa* and *M. arvensis* seemed to be less well-adapted, having lower dry-weight percentages. Weeds constituted more than 50% of the total dry weight in 2006, but this decreased to 23% in 2007 (Table 1), and therefore this legume mixture was considered to be successful in weed control.

Forage was well accepted by the sheep, as grazing in spring 2006 led to a biomass consumption of almost 42% of the herbage allowance (grazing vs. no grazing treatment, Table 1), but there were differences in the degree of consumption among species. Contrary to expectations, *M. arvensis* was rejected by livestock, and no grazing was detected in 2006, neither in field observation nor in the clipped samples (Table 1). However, in spring 2007 the same sheep flock appeared to show a high preference for this species (personal observation). The grazing time in spring 2006 may have been insufficient to activate the mechanisms of learning in food selection (Provenza *et al.*, 1992), but these mechanisms were activated in the following year. *Vicia ervilia* was consumed by sheep in spring 2006 (around 30%), although differences between the grazing and no-grazing treatments were not significant. *Medicago sativa* showed an unexpected performance, as it showed a higher percentage in the grazing treatment than in the no-grazing treatment in 2006. This must be due to uneven broadcast of seeds, as one of the grazing plots could have received more *M. sativa* seeds than others. In the field it could be observed that this species was heavily consumed. Finally, weed grazing was undetectable.

Self-reseeding was detected for every species and treatment. In fact, *M. arvensis* was clearly more abundant in 2007 than in 2006 (Table 1). The grazing period in 2006 decreased the self-



reseeding capability of *V. ervilia* in 2007, probably because grazing occurred during the flowering period, resulting in a lower number of pods than in the no-grazing treatment. *Medicago sativa* also increased its dry weight percentage in 2007, although this percentage was higher for the grazing than for the no-grazing treatment. The above-mentioned failure during the seeding process may have led to a higher number of seeds in one of the 'grazing-plots', and consequently the number of plants producing seeds was also higher for these plots in 2007.

Table 1. Mean biomass production and dry weight percentage ( $\pm$  SE) of the three sown species and of weeds.

	Biomass (kg DM ha <sup>-1</sup> )	<i>M. arvensis</i> (%)	<i>V. ervilia</i> (%)	<i>M. sativa</i> (%)	Weeds (%)
2006					
Grazing	849.12 $\pm$ 52.77 <sup>a</sup>	6.76 $\pm$ 2.72 <sup>a</sup>	25.09 $\pm$ 2.39 <sup>a</sup>	12.70 $\pm$ 2.05 <sup>ab</sup>	59.29 $\pm$ 6.96 <sup>a</sup>
No grazing	1456.87 $\pm$ 62.39 <sup>b</sup>	6.41 $\pm$ 2.76 <sup>a</sup>	35.44 $\pm$ 4.96 <sup>ab</sup>	3.80 $\pm$ 1.43 <sup>a</sup>	54.34 $\pm$ 5.45 <sup>a</sup>
2007					
Grazing	1959.70 $\pm$ 148.84 <sup>c</sup>	26.36 $\pm$ 5.00 <sup>b</sup>	21.80 $\pm$ 8.55 <sup>a</sup>	28.82 $\pm$ 13.62 <sup>b</sup>	23.02 $\pm$ 6.97 <sup>b</sup>
No grazing	1940.69 $\pm$ 39.95 <sup>c</sup>	15.05 $\pm$ 5.17 <sup>ab</sup>	45.56 $\pm$ 6.55 <sup>b</sup>	8.86 $\pm$ 2.71 <sup>ab</sup>	26.69 $\pm$ 3.87 <sup>b</sup>
Interaction	***	ns	ns	ns	ns

Different letters indicate significant differences within one column (Fisher-LSD test,  $P < 0.05$ ). Interaction between factors within each column (Grazing x Year) is indicated with ns (non significant), or \*\*\* ( $P < 0.01$ ).

## Conclusions

Our results indicate that this legume-cruciferae mixture can produce high yields of forage that can be consumed in spring and/or summer (the critical season for livestock feeding in arid and semiarid climates). Moreover, it may maintain the forage production for several years due to its high self-reseeding ability, and it can be successful in weed control. *Vicia ervilia* shows the best performance in yield production and self-reseeding ability. Therefore, it could be recommended as a useful species for marginal areas in semiarid and arid climates. Further studies and higher sample numbers are required to confirm our results.

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# Improved estimation of mountain grassland production by using local input data in a crop model

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## Abstract

The STICS crop model has been successfully tested in France to predict production of different crops and, since 2000, has also been used for grasslands at the regional scale by the French Ministry of Agriculture. Overestimation of grass production in highland conditions may be due to model functions or model parameters (using a 'mean' species in a multi-species community), or input data. The aim of this study was to explain this overestimation. Simulated aboveground biomass was compared with estimates made with field estimations over a seven-year period in the Vosges mountains (NE France, altitude: 250 to 1,300 m).

However, the use of accurate climatic data instead of regional values as input data in the STICS crop model still did not completely eliminate the overestimation. Introduction of the characteristics of the main species present, based on analysis of the species according to their functional traits, could enable better estimation of DM production of mountain grasslands. This result now needs to be confirmed especially by comparing it with the possible effects of other factors, like run-off, that influence the water balance and that may have been neglected up to now.

Keywords: grassland, crop model, mountain climate, functional traits, production

## Introduction

The STICS crop model has been successfully tested in France to predict production of different crops and has also been used at regional level for grasslands by the French Ministry of Agriculture. Overestimation of grass production in highland conditions could be due to uncertainties in climatic variables (= driving variables), or to model parameters (using a 'mean' species in a multi-species community), or to inadequate model functions. In this paper, we evaluate possible improvements to be obtained by using climatic variables and plant features as parameters in the model. Both climatic input data and several species-specific plant parameters that affect plant production are of great concern.

## Materials and methods

Plant parameters from the STICS model (Brisson *et al.*, 1998) were adapted for grassland cut at any stage of growth. A mean grass parameter set was used which was derived for intensive grass crops in lowland experiments (Ruget *et al.*, 2006). In addition to plant parameters, the model requires information on climate, soil and agricultural practices. The observed dataset contains field estimations made over a period of five years (2000 to 2004) in the main stock-breeding areas in the Vosges mountains (NE France, altitude: 250 to 1300 m). Estimates of aboveground biomass were based on observations of the number and weight of bales, or on estimates of how much the cattle consumed based on grazing duration and the mean daily dry matter eaten by a dairy cow. Permanent information on soil, climate and practices was collected as environmental patch features. These data were either not the same as those used in the model, or not at the same time scale as the input data required by the crop model. It was thus necessary to derive suitable data from measured data. Available water, pH and the C/N

ratio were observed, and field capacity, wilting point, depth and organic nitrogen are required for soil. In the same way year-to-year monthly mean values of climate variables (temperature, precipitation, global radiation, and potential evapotranspiration. ETP) were collected, and yearly daily values are needed. Year-to-year mean practices were collected (mean dates of first cut, number of cuts or mean fertilization amounts) and used as they were. Botanical composition was observed in each patch. As all French grassland species can be classified in four plant functional types (PFT), A, B, C and D, the patches were also classified in four PFTs as a function of the proportion of each species. The growth features of the PFT are well known (Table 1, Cruz *et al.*, 2003). The values of the Ellenberg indices (Ellenberg, 1991) were assigned for each species.

Table 1. Main features of the four functional types.

	Max prod.	Date of max prod.	Leaf life span	Digestibility	SLA and water content	Phenology
A	H	E	s	h	h	e
B	H	L	lg	m	h	
C	Low	L	lg	low	low	l
D	Low	VI	vlg	very low	low	

Prod: production, h: high, , e: early, l: late, vl: very late, s: short, lg: long, vlg: very long, m: medium.

A number of alternative daily climate datasets were derived from the available information (Table 2). The first used the climate at a single location (Colmar, series 1), the second (series 2) used the local and daily values known for each patch, obtained with correction factors from mean monthly values of the Aurelhy interpolation method (Benichou and Le Breton, 1986). The third (series 3) used classical meteorological relations (temperature with height, ETP with formulas and precipitation at the nearest stations to eight nearby locations), and for the fourth and final (series 4), we used a combination of the best estimation for each variable. Global radiation was the same in all datasets because of the low variations between values at locations caused by the Aurelhy method. To evaluate the methods, we compared the results of six input sets, using only statistical criteria (RMSE, root mean square errors and its systematic and unsystematic components).

Table 2. Composition of the 4 meteorological datasets as a function of the origin of the meteorological data.

Variable	'Unique' 1	Aurelhy interpolation 2	STICS calc and local measure 3	STICS calc and Aurelhy 4
T	Colmar	monthly Aurelhy diff	STICS calc with altitude	STICS calc with altitude
ETP	Colmar	Colmar	calc using simple formulas	calc using simple formulas
RR	Colmar	monthly Aurelhy diff	near stations	monthly Aurelhy diff
Rg	Colmar	Colmar	Colmar	Colmar

(T: temperature, ETP: potential evapotranspiration, RR: precipitation, Rg: solar radiation, calc: calculation, diff: differences, measure: measurements).

## Results and discussion

Classifying species using Ellenberg indices or plant functional types (PFT) gives approximately the same results. The good agreement between the classification of species obtained with PFT typology and Ellenberg indices (AFC) is one of the main results of our study: most of the species are grouped together in the same groups (classes). Moreover, there is an altitudinal distribution of the species and PFT, with A and B species at low altitudes, C and D at high altitudes. The features of both main species present in the Vosges mountains (B and D), led us to give new values to two parameters, driving the leaf growth rate and the crop production through light use efficiency. We thus constructed two plant files, one featuring species with a high leaf growth rate and high production (two parameters) and the other with half values for both parameters.

Climatic series differed considerably for rain between rain shadow valley values, station values and Aurelhy values (higher maximal values at high altitudes). The ETP values differed when they were estimated using simple formulas introducing the effect of altitude on temperature, instead of unique or Aurelhy values (more or less similar).

Combining some among the four climatic series with both plant parameter files led to six main simulation sets, whose results are reported below in Table 3.

Table 3. Statistical results of tested combinations of plant features and climate calculations.

Climate	1	4	1	2	3	4
Plant features	prairie	prairie	BD	BD	BD	BD
RMSE	1.6	2.57	1.31	1.52	1.58	1.34
RMSEs	0.83	2.06	0.82	1.2	1.13	1.00
RMSEu	1.37	1.54	1.02	0.92	1.11	0.90

Prairie: the unique original plant file, BD, two plant files, i.e. consideration of plant functional types.

1 to 4: the numbers of climatic sets (table 1).

The best results came from the use of the plant files featuring the PFT and as climate data, either unique climate, or climate using Aurelhy rain and altitudinal variations in temperature and ETP. Paradoxically, the correct value of statistical criteria in the configuration of a unique lowland climate (1) is probably due to the serious water stress that occurs with the rain shadow climate. The last simulation (climate 4 and plant BD) gave the lowest dispersion (RMSEu) and can be regarded as the best configuration.

The main conclusion is the interest of taking plant functional types into account. Moreover, the quality of estimations is highly dependent on the water balance. This means that, in such conditions, both rain and ETP must be well assigned.

## Conclusion

The classification of species obtained using Ellenberg indices (without correspondence with the model parameters) and with the PFT (with correspondence) led to the same groups of plants. This meant the species could be gathered in two main groups and the plant parameters could be attributed for each existing group (B and D). The introduction of the characteristics of the main species present, based on analysis of the species according to their functional traits, enabled better estimation of DM production of mountain grasslands. However, only one trial was conducted that showed the ability of the model to represent functional types, but we did not study either all the functional types or all the plant parameters involved. Climate also has a major effect on production. The construction of a daily climate database using different methods for estimating each variable gave the best series when using each method in their best domain: Aurelhy for rain, and altitudinal calculation for temperature and ETP.

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# Influence of mineral fertilization on production and quality of forage in a Sardinian pasture

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## Abstract

A three-year trial was carried out on a Sardinian hill pasture to study the influence of mineral fertilization on forage production and quality. Three different fertilizers were tested in single and combined forms to distribute: 100 kg N ha<sup>-1</sup>, 57 kg P ha<sup>-1</sup> and 108 kg K ha<sup>-1</sup>. Phosphorus increased total forage yield in the first and third years and the production of legumes in all three years. It also changed the chemical composition of forage in the second year with a significant increase in crude protein, calcium and phosphorus and a reduction in NDF and ADF content. Nitrogen always decreased legume production and it improved forage yield in the first year only. Moreover, it caused a higher level of NDF and a lower level of calcium and phosphorus in the last two years. Potassium affected forage production in the first year only. Phosphorus fertilization increased its availability in the soil, reaching an adequate level in the last year.

Keywords: forage yield, forage quality, mineral fertilization, legumes

## Introduction

Sardinian pastures are characterized by a wide range of pedoclimatic conditions which influence forage production and quality. The annual meteorological trend plays an important role in the seasonal distribution of pasture yield (Bullitta *et al.*, 1987) especially in autumn and winter. Moreover, human activity can influence the availability of pasture herbage through stocking and agronomic techniques such as over sowing, irrigation and fertilization (Arangino, 1990). Mineral fertilization is a simple and immediate method for increasing forage production and for changing the floristic composition of the pastures. The results depend on several environmental and management factors, such as natural mineral availability in the soil, seasonal rainfall and temperature, conditions of waterlogging, stocking rate, etc.

## Materials and methods

The trial was carried out from 2004 to 2006 in a hilly pasture area in central Sardinia characterized by periods of waterlogging in the autumn and winter season. The climate is classified as mesothermal-humid with average annual rainfall above 1,000 mm and average temperatures varying from 4 °C to 32 °C. The meteorological trend over the three years showed lower than average values of temperature and rainfall. The clay sub-acid soil was poor in available phosphorus and exchangeable potassium and had a sufficient level of total nitrogen. In a split-split plot experimental design, three different mineral fertilizers, ammonium nitrate, triple superphosphate and potassium sulphate were tested in order to distribute every year, in single or combined form, 100 kg ha<sup>-1</sup> of N, 57 kg ha<sup>-1</sup> of P and 108 kg ha<sup>-1</sup> K.

Two herbage samples were collected from areas of 0.5 m<sup>2</sup> for each plot to evaluate forage production and then partitioned into floristic groups in order to determine their botanical composition. Due to waterlogging during the cold season, forage yield was determined in the spring period; it was possible to evaluate autumn production in the first year only. In spring of



2005 and 2006 chemical fractions, crude protein (CP), neutral detergent fibre (NDF) acid detergent fibre (ADF), acid detergent lignin (ADL), calcium and phosphorus, were determined to evaluate the quality of forage yield. In the autumn of the last year, soil chemical characteristics were also monitored. All data were analysed by ANOVA.

## Results and discussion

Phosphorus fertilization increased forage yield in the first and third year and also the percentage of legumes in the second and third year (Table 1). The legume yields, in years 2004, 2005 and 2006 respectively, were increased significantly from 1,267 to 1,850 kg DM ha<sup>-1</sup>, from 533 to 1,583 kg DM ha<sup>-1</sup> and from 173 to 376 kg DM ha<sup>-1</sup>. Chemical composition of forage in the spring of 2005 displayed a significant increase in crude protein, calcium and phosphorus and a reduction in NDF and ADF content (Table 2). The low presence of legumes could be the reason for the not-significant difference in most chemical fractions of forage in 2006.

Table 1. Total dry matter (DM) forage yield and percentage of legumes and grasses in each of the three years.

	2004			2005			2006		
	DM kg ha <sup>-1</sup>	Legumes %	Grasses %	DM kg ha <sup>-1</sup>	Legumes %	Grasses %	DM kg ha <sup>-1</sup>	Legumes %	Grasses %
N <sub>0</sub>	6,465	29	69	4,539	38	61	4,682	10	87
N <sub>1</sub>	7,301	10	86	4,878	8	91	4,900	3	94
	*	**	**	ns	**	**	ns	*	ns
P <sub>0</sub>	6,212	17	79	4,671	12	87	4,421	5	93
P <sub>1</sub>	7,553	21	76	4,745	34	65	5,160	8	88
	**	ns	ns	ns	**	**	**	*	*
K <sub>0</sub>	6,499	20	77	4,599	23	77	4,765	5	91
K <sub>1</sub>	7,266	19	78	4,818	24	75	4,816	7	90
	*	ns	ns	ns	ns	ns	ns	ns	ns
P x N	ns	ns	ns	ns	*	ns	ns	ns	ns
P x K	ns	**	*	ns	ns	ns	ns	ns	ns

\* = values differ significantly at  $P < 0.05$ , \*\* = values differ significantly at  $P < 0.01$ , ns = not significant. (N<sub>0</sub>, P<sub>0</sub>, K<sub>0</sub> = no fertilized; N<sub>1</sub>, P<sub>1</sub>, K<sub>1</sub> = fertilized).

Table 2. Chemical composition of forage in spring season in the last two years.

	NDF		ADF		ADL		CP		Ca		P	
	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006	2005	2006
	g kg <sup>-1</sup>		g kg <sup>-1</sup>		g kg <sup>-1</sup>		g kg <sup>-1</sup>		g kg <sup>-1</sup>		g kg <sup>-1</sup>	
N <sub>0</sub>	483	449	330	289	41	35	126	146	7.5	4.4	2.4	3.2
N <sub>1</sub>	560	484	377	311	39	33	118	140	4.0	3.8	2.1	3.0
	*	*	ns	ns	ns	ns	ns	ns	*	ns	*	*
P <sub>0</sub>	552	472	367	301	41	33	105	144	3.9	3.7	1.8	2.6
P <sub>1</sub>	491	461	341	298	39	35	140	142	7.6	4.5	2.6	3.6
	**	ns	**	ns	ns	ns	**	ns	**	**	**	**
K <sub>0</sub>	524	467	354	298	42	35	124	142	5.8	4.3	2.2	3.2
K <sub>1</sub>	519	466	354	301	38	33	120	145	5.7	3.9	2.2	3.0
	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
P x N	**	ns	**	ns	ns	ns	**	ns	**	ns	**	ns
P x K	ns	ns	ns	ns	ns	ns	ns	*	ns	ns	ns	ns

\* = values differ significantly at  $P < 0.05$ , \*\* = values differ significantly at  $P < 0.01$ , ns = not significant. (N<sub>0</sub>, P<sub>0</sub>, K<sub>0</sub> = no fertilized; N<sub>1</sub>, P<sub>1</sub>, K<sub>1</sub> = fertilized).

Nitrogen always decreased the percentage of legumes in pasture, and increased grasses in 2004 and 2005; it improved total forage production in the first year only. Moreover, it

produced a significantly higher level of NDF, and a lower level of phosphorus, in the second and third years, and a lower level of calcium in the second year only.

Potassium affected the production of forage in 2004 only, and did not influence its chemical composition.

An interesting finding was the significant interaction between phosphorus and potassium in the first year, and phosphorus and nitrogen in the second year, in terms their effects on the proportion of legumes (Table 3). Potassium and nitrogen fertilizers, combined with phosphorus, negatively affected the increase in legumes that occurred under single phosphorus fertilization. In the same way there was a negative interaction in 2005 between phosphorus and nitrogen in terms of most of the chemical concentrations (Table 2).

Table 3. Phosphorus-potassium and phosphorus-nitrogen interaction in the legumes presence.

	2004		2005
	Legumes (%)		Legumes (%)
P <sub>0</sub> K <sub>0</sub>	14 B	P <sub>0</sub> N <sub>0</sub>	21 b
P <sub>1</sub> K <sub>0</sub>	26 A	P <sub>1</sub> N <sub>0</sub>	55 a
P <sub>0</sub> K <sub>1</sub>	20 AB	P <sub>0</sub> N <sub>1</sub>	4 d
P <sub>1</sub> K <sub>1</sub>	17 AB	P <sub>1</sub> N <sub>1</sub>	13 c

Means with the same capital or small letters are not different, respectively, for  $P \leq 0.01$  and  $P \leq 0.05$ .

Finally, phosphorus fertilization significantly increased the level of available phosphorus in the soil, from 7.0 mg kg<sup>-1</sup> to 17.1 mg kg<sup>-1</sup>. The other fertilizers did not affect the principal chemical characteristics of the soil.

## Conclusions

The trial results confirm the importance of phosphorus fertilization in environmental conditions characterized by long periods of waterlogging. It improved yield and pasture quality, raising the available phosphorus in the soil to a sufficient level. In cases where there is a good natural level of nitrogen in the soil, nitrogen fertilization should not be necessary because it might heavily shift the floristic balance towards grass species. Moreover, it might interfere with the positive action of phosphorus on legumes. Lastly, the use of potassium showed no advantage despite its low content in the soil.

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# Grassland productivity and consumption under different tree cover in the Aysén region, Patagonia, Chile

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## Abstract

Herbaceous vegetation under different tree cover has been studied in the Aysén region in the northern Patagonia of Chile, in order to test the feasibility of silvopastoral agroforestry in this region. Within a moderately altered deciduous forest of Lenga (*Nothofagus pumilio*), grassland productivity and consumption by cattle was estimated in 400 m<sup>2</sup> plots with different degrees of tree cover, from 0-80%. In each plot, under and between tree crown patches were compared. No differences were observed in grassland production between full-open grassland and plots with different tree cover. However, with increasing tree cover, herbaceous production and consumption decreased under tree crowns, but this decrease was compensated for by an increase in productivity between tree crowns. This resulted in similar average pasture production in open grassland and with trees, across the range of cover. Taking into account the trade off mentioned above, it appears that maintaining Lenga forest on pasture land, with tree cover ranging from 50 to 75%, yields optimal productivity for cattle in this region, and allows production of valuable timber as well. Thus, silvopastoral agroforestry is an attractive option for this region.

Keywords: silvopastoral agroforestry, canopy, cattle, forest fragmentation, grassland management, *Nothofagus pumilio*, plant biomass

## Introduction

Foresters and agronomists have historically managed forest and grassland resources independently in the Aysén region of northern Patagonia of Chile, both with different approaches and objectives. In this region, over five million ha were transformed from forest to grasslands in the last century, as a consequence of human-set fires (Donoso and Lara 1996, Veblen *et al.*, 1996, Cruces *et al.*, 1999, Silva *et al.*, 1999). Soil erosion and forest loss count as major negative consequences for environment and socio-economical development. There is a need for scientific research aimed at setting the ecological basis of a silvopastoral management model. There is also a need for scientific research to test the feasibility of agroforestry in this region, to allow pasture and animal production without compromising forest preservation. This study attempts to determine the direct influence of tree cover on grassland productivity and consumption by cattle. To achieve this we analysed above-ground herbaceous biomass production of grasslands along a gradient of forest fragmentation.

## Material and methods

The study was conducted in a moderately altered Lenga (*Nothofagus pumilio* [Poepp. et Endl.] Krasser) forest in the Patagonian region of Aysén, Chile (44 to 48° S). The climate is cold and arid with low precipitation and strong winds. Vertical projection of tree crowns on the ground was used as an expression of tree cover. We distributed 15 plots (20 x 20 m) in five classes of tree cover (three plots per class) from open grassland to dense forest: 0%, 1 to



25%, 25 to 50%, 50 to 75%, and over 75%. In each plot two patches were considered, under and between tree crowns. In each patch we determined herbaceous production and consumption by cattle during the last growing season (December to March 2006-2007). Above-ground herbaceous biomass was evaluated monthly (four times) by cuttings at random four subplots (100 x 50 cm) at ground level, using an electric shearing handpiece. Half of the subplots were excluded from grazing with the use of cages, which we moved within the same sampling area after cutting. Exclusion cages were used to determine net primary production (NPP) as the biomass difference between two consecutive times. For each time period, consumption was estimated as the difference of biomass inside and outside the cage at the end of the period. Temporal variation in NPP and consumption was averaged for the three time periods.

## Results and discussion

Considering the two patches (under and between tree crowns) jointly, only the plots representing dense forest (over 75%) tended to have a NPP slightly lower than that of the other cover classes, although no significant differences were detected (Tukey, test of means comparison,  $P > 0.05$  in all cases) (Figure 1a).

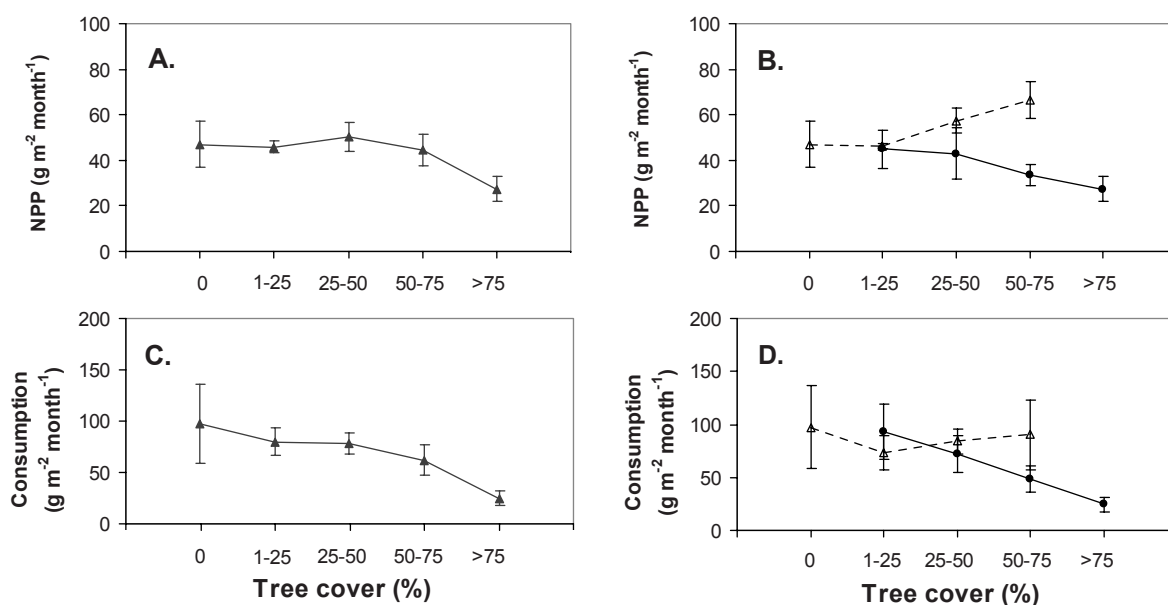


Figure 1. Effects of tree cover on mean values of net primary production (NPP, a), and consumption by cattle (c). In b) and d) values under and between tree crowns are shown separately (continuous and dashed line, respectively).

The tree crown had an effect on NPP which was evident with tree cover higher than 25% (Figure 1b). Above this value net primary production diverged, increasing in patches located between tree crowns and decreasing in those located under the trees. Mean differences between both situations were only significant in the tree cover class from 50 to 75% ( $t$  test = 3.8;  $P = 0.007$ ). This resulted in similar average NPP in open grassland and with trees, across the range of cover (Figure 1a).

Consumption values decreased as tree cover increased (Spearman test,  $r = -0.349$ ,  $P = 0.024$ ; Figure 1c). This tendency was mainly associated with differential consumption under and between tree crowns (Figure 1d). In the former, consumption tended to decrease (Spearman

test,  $r = -0.62$ ,  $P = 0.003$ ), while in the latter it did not vary with tree cover (Spearman test,  $r = 0.097$ ,  $P = 0.676$ ).

These results support the implementation of a silvopastoral agroforestry system with dispersed trees (Lenga) in northern Patagonia, similar to 'Dehesas' in Spain (Olea and San Miguel, 2006) or the 'Espinales' in the Mediterranean area of Chile (Ovalle, 1986; Ovalle *et al.*, 1990). Our preliminary results suggest that, in these systems, the presence of dispersed trees would not reduce grassland productivity; indeed, this appears to increase in gaps between tree crowns. Additionally, the presence of trees in a pasture matrix would contribute to preserving naturalistic values (biodiversity conservation, additional habitat for livestock, among others; De Miguel *et al.*, 1988).

## Conclusions

In silvopastoral systems different tree covers determine variations in grassland productivity and consumption by cattle. Grassland productivity presents little variation in different scenarios of forest fragmentation, from full-open areas to relatively dense forests. However, this uniformity of values is acquired by a divergent pattern of net primary production under and between the tree crowns. Herbage intake is similar in gaps between trees, regardless of the degree of fragmentation, whereas it decreases under the tree crowns with increasing tree cover. These results would support the implementation of a silvopastoral management system as an alternative to the dominant open-pasture model in this region.

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# **Are herbage yield and yield stability affected by plant species diversity in sown pasture mixtures?**

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## **Abstract**

A tenet of plant biodiversity theory in grasslands is that increased diversity contributes to the stability of ecosystems. In managed grasslands, such as pastures, greater stability of herbage production as a result of increased plant species diversity would be beneficial. In this study, I combined historical data from pasture mixture experiments conducted during the 1930s to 1960s in the USA along with more recent data from my laboratory to determine the relationships among herbage yield, stability of yield [measured as the relative standard deviation (RSD)], and diversity measures [Shannon diversity index (H), species richness (S), and evenness, (J)]. In nearly all studies, mixtures yielded more herbage than monocultures and the RSD of yield across years was less for mixtures than monocultures. There was a wide variation in the derived relationships within mixtures, however, and in many instances there was no relationship between herbage yield or yield stability and the complexity of the mixture.

Keywords: plant species diversity, evenness, species richness, yield stability

## **Introduction**

The use of forage mixtures in pastures has a long history. Taking their cue from the plant diversity of natural grassland communities, some farmers often plant complex mixtures of grasses and legumes (Sanderson *et al.*, 2007) because they believe that maintaining a highly diverse botanical composition in pastures benefits yield stability, persistence, and productivity. Re-examining previous grassland studies in the light of new ecological concepts can be a useful and efficient way to glean new information about forage mixtures. I used three studies from the 1930s, 1940s, and 1960s along with current research to examine relationships among herbage yield, yield stability, and components of plant species diversity.

## **Materials and methods**

Data from three historical studies were used in the analysis. The studies were chosen because (i) a large number of mixtures were compared, (ii) details on the proportions of individual species in each mixture were available, and (iii) data are reported individually by year. The first study included 50 different single and multiple species combinations of cool-season grasses and legumes (1, 2, 3, 4, or 7 species) for yield under clipping for three years (1933, 1934, and 1935) in Connecticut, USA (Brown and Munsell, 1936). This report also included data on the legume proportions of the herbage harvested from the plots each year. The second study was comprised of three separate experiments, which in total compared 93 combinations of cool-season grasses and legumes (1 to 7 species) in both clipped and grazed plots under irrigation during 1944 to 1949 in Utah, USA (Bateman and Keller, 1956). Data in the Utah study were reported on green fresh weight basis. The third study included 38 mixtures of cool-season grasses and legumes grown in Pennsylvania during 1967 to 1969 (Washko *et al.*, 1974). A fourth study was a recent experiment that compared 20 mixtures and monocultures

under grazing at University Park, Pennsylvania during 2002 to 2004 (Hall, 2006; Deak *et al.*, 2007).

For each study, the number and relative proportions of each species planted in the mixtures were used to calculate the Shannon diversity index [ $H = -(\sum(\rho_i \ln \rho_i))$ , where  $\rho_i$  is the proportion of the total number of species  $i$  expressed as a proportion of the total number of species for all species in the sample] and an evenness index ( $J = H/H_{\max}$ ) (Magurran, 1988). The dry matter yield of each mixture in each study was then related to  $S$  (species richness or number of species in the mixture),  $H$ , and  $J$  via correlation and regression procedures with the Statistical Analysis System (SAS Inst., 1999). Additionally, the relative standard deviation (RSD; standard deviation divided by the mean and expressed as a percentage) of dry matter yield across years was calculated and then related to  $H$ ,  $S$ , and  $J$ .

## Results and discussion

There were no significant relationships between herbage yield or RSD with diversity indices in the Connecticut study (Table 1). Herbage yield was positively correlated with  $S$ ,  $H$ , and  $J$  in the Utah study; however, there was no relation with RSD. In the Pennsylvania study (Washko *et al.*, 1974), herbage yield was positively related with  $S$  and  $H$  but not  $J$ , whereas RSD was positively related to  $J$ . In the recent Pennsylvania study (Deak *et al.*, 2007), herbage yield was positively related to  $S$ . Table 2 summarizes the average yield and RSD for the studies. In each study, the RSD was highest for monocultures compared with the mixtures. Within mixtures, however, there was no consistent relationship between the number of species and RSD. This may indicate that species identity and composition of the mixture may be more important determinants of yield and yield stability than simply the number of species combined in a mixture as suggested by Deak *et al.* (2007). The legume proportion data from the Connecticut study support this premise. In that study, herbage yield was positively related ( $r^2 = 0.33$ ,  $P < 0.05$ ) to the proportion of legume in the harvested herbage, indicating an effect of mixture composition.

Table 1. Relationships ( $r^2$ , coefficient of determination) of herbage yield and yield stability (relative standard deviation, RSD) with diversity indices in four studies conducted in the USA.

Diversity Index <sup>1</sup>	Connecticut <sup>2</sup>		Utah <sup>3</sup>		Pennsylvania <sup>4</sup>		Pennsylvania <sup>5</sup>	
	Herbage yield	RSD	Herbage yield	RSD	Herbage yield	RSD	Herbage yield	RSD
$S$	ns <sup>6</sup>	ns	0.13**	ns	0.25**	ns	0.34**	ns
$H$	ns	ns	0.15**	ns	0.23*	ns	ns	ns
$J$	ns	ns	0.05*	ns	ns	0.21*	ns	ns

<sup>1</sup> $S$  = species richness;  $H$  = Shannon diversity index;  $J$  = evenness; <sup>2</sup>Brown and Munsell (1936), <sup>3</sup>Bateman and Keller (1956), <sup>4</sup>Washko *et al.* (1974), <sup>5</sup>Deak *et al.* (2007) and Hall (2006), <sup>6</sup>ns = not significant; \*, \*\* = significant at  $P < 0.05$  and  $0.01$ , respectively.

Table 2. Yield and relative standard deviation (RSD) for several mixtures of grasses and legumes in four studies conducted in the USA. Data for the Utah study are expressed as green fresh weight of forage. Other data are on a 100% dry matter basis.

Species in mix	Connecticut <sup>1</sup>		Utah <sup>2</sup>		Pennsylvania <sup>3</sup>		Pennsylvania <sup>4</sup>	
	Yield	RSD	Yield	RSD	Yield	RSD	Yield	RSD
	Mg ha <sup>-1</sup>	%	Mg ha <sup>-1</sup>	%	Mg ha <sup>-1</sup>	%	Mg ha <sup>-1</sup>	%
1	1.5	57.3	28.4	19.3	8.1	15.5	5.8	18.7
2	2.5	28.1	28.7	17.4	8.3	12.3	8.6	14.5
3	2.3	19.0	29.2	16.5	8.5	5.1	8.4	10.4
4	1.7	33.0	30.0	16.4	9.2	10.8		
5			30.7	12.0				
6	2.6	41.8	33.6	11.4	9.0	6.7	9.9	12.3
7	2.6	36.6	33.6	10.1				
9							9.6	10.1

<sup>1</sup>Brown and Munsell (1936), <sup>2</sup>Bateman and Keller (1956), <sup>3</sup>Washko *et al.* (1974), <sup>4</sup>Deak *et al.* (2007) and Hall (2006).

## Conclusions

Monocultures consistently had the highest RSD of herbage yield across years compared with forage mixtures. Within forage mixture, however, there was no consistent relationship between herbage yield or yield stability (RSD) and measures of species diversity. Species identity and composition of forage mixtures may be more important determinants of herbage yield than simply the number of species.

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# SDS-protein and isozyme markers of alfalfa genotypes and their offspring in response to drought and salinity stresses

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## Abstract

Fifteen alfalfa landraces from different regions in Egypt were used to study the effect of drought and salinity stresses depending on some yield-related traits. Two landraces were chosen as drought/salinity-tolerant and drought/salinity-sensitive. SDS-protein and isozymes (peroxidase, esterase and acid phosphatase) were used to identify some biochemical markers associated with drought and salinity tolerance for these two landraces and their F<sub>1</sub> and F<sub>2</sub> seeds. The results indicated that SDS-PAGE of water-soluble proteins exhibited a maximum number of 29 bands. Two of these bands could be considered as positive markers associated with drought- or salinity-tolerance, respectively. Peroxidase and acid phosphatase isozymes showed differential responses with respect to drought and salt tolerance. Esterase did not exhibit any biochemical marker for drought and salinity tolerance in this study.

Keywords: alfalfa, drought, salinity, SDS-protein, isozymes

## Introduction

Salinity and drought stresses are the main obstacles to increasing the yield in newly reclaimed and semi-arid lands. In Egypt, alfalfa is grown mainly in these lands, which suffer from salinity and drought. Electrophoretic techniques have been used to identify and quantify protein and isozyme polymorphisms. The altered expression of specific genes correlate with changes in the environment, as changes in expression of genes may be involved in adaptation and could be used as molecular markers for salt and drought stresses. The aims of this study were: 1) to evaluate 15 landraces under salt and drought conditions and to select the most tolerant and most sensitive ones according to some yield-related traits, and 2) to identify molecular genetic markers associated with salt and drought tolerance using SDS-protein and isozymes.

## Materials and methods

Seeds of 15 alfalfa landraces were sown in a sand culture experiment. The plants were divided into three random groups, T<sub>1</sub> (6,000 ppm NaCl), T<sub>2</sub> (drought by withholding irrigation for 30 days) and T<sub>3</sub> (control which was irrigated weekly with Hoagland solution). The two contrasting landraces, and their F<sub>1</sub> and F<sub>2</sub> seeds, were sown in a sand culture experiment for salinity and drought. Data were recorded on samples for plant height (cm), number of branches, leaf fresh weight (g), stem fresh weight (g), leaf/stem ratio for fresh weight, and total fresh and dry forage yield (g). The collected data were statistically analysed according to Snedecor and Cochran (1969). SDS-PAGE was performed on water-soluble and non-soluble leaf protein fractions according to the method of Laemmli (1970) modified by Studier (1973). Isozyme fractions were separated according to Stegemann *et al.* (1985).



## Results and discussion

Based on the yield-related traits, plants that showed the highest frequency under drought or salinity were selected as the most tolerant bulk, while plants that showed the lowest frequency were chosen as the most sensitive bulk (data not shown). SDS-protein and isozyme electrophoretic patterns of the two parents, F<sub>1</sub> and F<sub>2</sub> bulk alfalfa under drought, control and salt treatments, respectively are shown in Figure 1. Electrophoresis of water-soluble protein fractions exhibited a maximum number of 29 bands (Figure 1a). None of the bands exhibited a specific trend for drought tolerance except band No. 26 (about 15.8 KDa) which was present in the tolerant parent and F<sub>2</sub> tolerant under drought, but it was absent in the sensitive parent and F<sub>2</sub> sensitive. Therefore, this band could be considered as a positive marker associated with drought tolerance in this study. Also, under salinity conditions, there was a band (No. 24, about 19.06 KDa) present in the tolerant parent and F<sub>2</sub> tolerant under salinity, but it was absent in the sensitive parent and F<sub>2</sub> sensitive. Therefore, this band could be considered as a positive marker associated with salinity tolerance in this material. These results are in agreement with Fahmy *et al.* (1992).

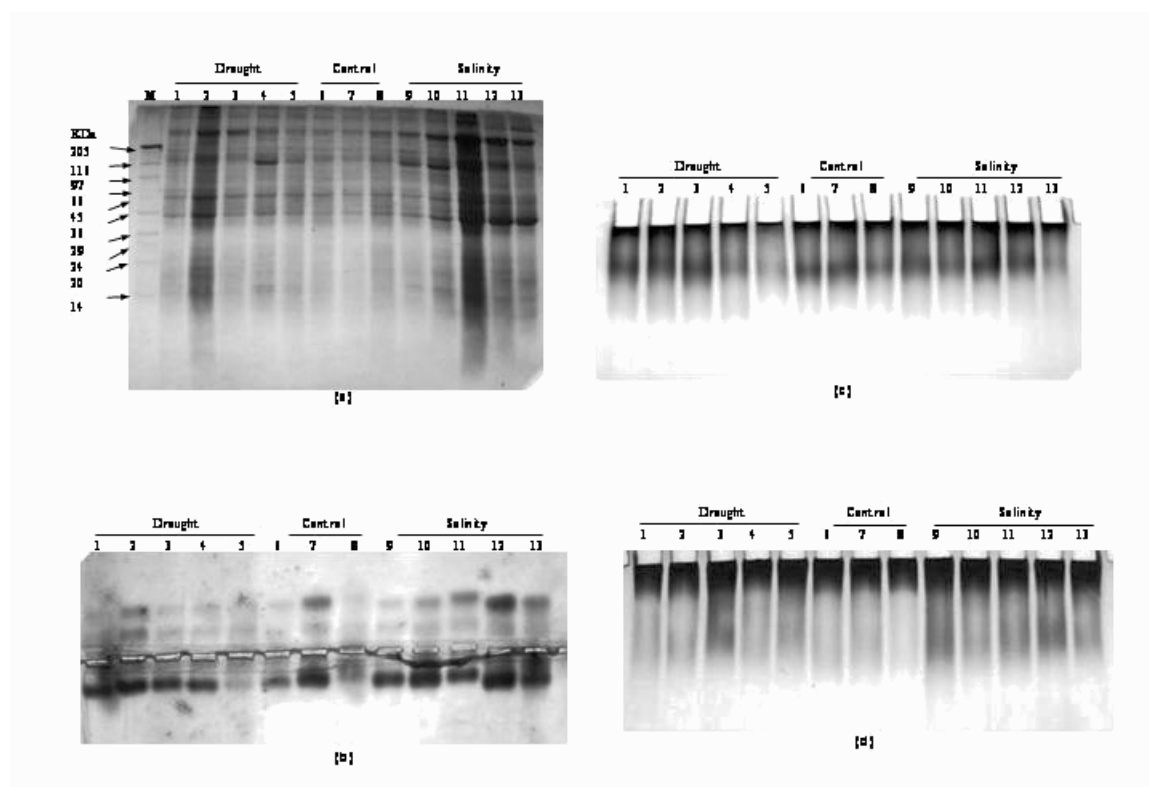


Figure 1. Electrophoretic patterns of SDS-PAGE profiles of alfalfa leaf water-soluble proteins (a), peroxidase (b), esterase (c) and acid phosphatase (d) for the two alfalfa parents, their F<sub>1</sub>, most tolerant F<sub>2</sub> and most sensitive F<sub>2</sub> bulk.

- |  |  |   |
|--|--|---|
| 1 = The drought tolerant parent                    | 2 = The drought sensitive parent                 | 3 = F <sub>1</sub> under drought                  |
| 4 = The bulk of F <sub>2</sub> drought tolerant    | 5 = The bulk of F <sub>2</sub> drought sensitive | 6 = The tolerant parent under control             |
| 7 = The sensitive parent under control             | 8 = F <sub>1</sub> under control                 | 9 = The salinity tolerant parent                  |
| 10 = The salinity sensitive parent                 | 11 = F <sub>1</sub> under salinity               | 12 = The bulk of F <sub>2</sub> salinity tolerant |
| 13 = The bulk of F <sub>2</sub> salinity sensitive | M: Molecular weight standard                     |   |

The results of electrophoretic patterns of peroxidase isozyme showed a total number of two bands appearing in the anodal direction and two bands towards the cathodal direction (Figure 1b). The esterase isozyme exhibited a total number of three bands which did not appear in all leaf



samples (Figure 1c). The activity of bands (intensity) increased under stresses (drought and salinity) but esterase isozyme patterns did not give remarkable markers to rely on for the discrimination between tolerant and sensitive F<sub>2</sub> under stresses. Acid phosphatase isozymes showed a maximum number of five bands (Figure 1d). Under drought, acid phosphatase isozyme patterns did not give clear-cut markers for the discrimination between drought-tolerant and drought-sensitive, but enzyme activity increased in drought compared with control. Under salinity, bands 3 and 4 could be considered as positive molecular markers for salt stress. These results are in agreement with Abdel-Tawab *et al.* (1997) and Rashed *et al.* (2001).

## Conclusions

The patterns of SDS-proteins and isozymes showed differential responses with respect to drought and salt tolerance. SDS-protein could be considered as a positive marker associated with stress tolerance. The activity of isozymes had increased under stresses compared with the control.

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# Productivity and quality of perennial ryegrass (*Lolium perenne* L.) in western Lithuania

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## Abstract

The productivity and quality of perennial ryegrass (*Lolium perenne* L.) pure stands were investigated in temporary grasslands in western Lithuania. The varieties differed in origin, ploidy level, and maturity. Under the agroecological conditions of western Lithuania perennial ryegrass swards produced a dry matter (DM) yield of 5.70-6.36 t ha<sup>-1</sup>. On average, ryegrass accounted for 89.2-92.9% of the sward DM matter yield. The coefficient of variation, ranging from 5.9 to 9.7%, suggests that the percentage of ryegrass cultivars Recolta, Respect and Meltra in the DM yield was the most stable in this respect. Crude protein content was similar in all the ryegrass varieties tested. The content of crude fibre depended on perennial ryegrass maturity group and leafiness. Significant differences in crude fibre content were identified when comparing the early-maturing varieties Sodre and Merlinda with medium-early or late-maturing foreign varieties. Leafier ryegrass varieties accumulated less crude fibre.

Keywords: perennial ryegrass, productivity, herbage quality

## Introduction

Perennial ryegrass (*Lolium perenne* L.) is distributed in countries with a maritime, mild climate, and widely used due to its high productivity (Nekrošas, 2002) high forage quality and digestibility (Wilkins, 1997). Cultivated plant species characterized by a high feeding value are more demanding in terms of nutrients and moisture than those characterized by a poor feeding value, and which are able to better adapt to adverse growing conditions. The varieties of perennial ryegrass with the highest ecological stability can spread over a wider ecological area (Katova *et al.*, 2007). Perennial ryegrass has been cultivated in Lithuania for a long time; however, it is not very widely distributed due to its poor overwinter survival and damage done to it by spring frosts and spring mould. Under the Lithuanian climate perennial ryegrass is not persistent, and its dry matter (DM) yield declines dramatically in the second year of use (Lemežienė *et al.*, 2004). The aim of this study was to compare perennial ryegrass varieties differing in origin, ploidy level and maturity in western Lithuania.

## Materials and methods

Nine varieties of perennial ryegrass differing in origin, ploidy level, and maturity group were studied at the Vezaiciai Branch of the Lithuanian Institute of Agriculture (western Lithuania). The soil of the experimental site is orthieutric albeluvisol (ABe-o), with pH 4.9-5.2, available P<sub>2</sub>O<sub>5</sub> content in the range 117-214 mg kg<sup>-1</sup>, and K<sub>2</sub>O of 173-211 mg kg<sup>-1</sup>. The trials were set up in 2000. The net plot size was 8.4 m<sup>2</sup>, arranged in one band with four replications. The swards were used for three years. In the first, second and third year of use the swards were fertilized with 120 kg N ha<sup>-1</sup>, 60 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> and 60 kg ha<sup>-1</sup> of K<sub>2</sub>O. Three cuts were taken per vegetation at the ear-emergence stage. Plant chemical composition was estimated by the following methods: nitrogen (N) after Kjeldhal, crude protein (CP) according to the amount of N x 6.25, and crude fibre (CF) by the Kurschner-Hanek method. The analyses of CP and CF were performed on all cuts of all years. The leafiness was determined by structural analysis.

The experimental data were computed with ANOVA and correlation-regression analyses according to Tarakanovas (1999).

Meteorological conditions were different in the different vegetation periods. The spring and summer of the year 2000 were warm and dry. In 2001 the plant growing season was rainy (rainfall 642.9 mm, which was by 126.4 mm more than long-term mean) and warm. In the spring of 2002 warm and dry weather prevailed. At the beginning of summer there was sufficient warmth and moisture for the development of perennial ryegrass, and in August with prevailing dry weather and declining moisture reserves, the conditions for herbage growth were only satisfactory. The drought lasted until the second ten-day period of September. In the spring and summer of 2003, except for July, hydrothermal conditions were favourable for the development of perennial grasses.

## Results and discussion

Averaged data indicate that under the agroecological conditions of western Lithuania the swards of perennial ryegrass produced a DM yield of 5.70-6.36 t ha<sup>-1</sup> (Table 1). Ryegrass accounted for 89.2-92.9% of the total DM yield of the sward. The least variation of this indicator was recorded for the varieties Recolta, Respect, Meltra and Merlinda (coefficient of variation from 5.9 to 10.7%).

Table 1. DM yield and quality parameters of different cultivars of perennial ryegrass.

Cultivar,		DM yield (t ha <sup>-1</sup> )				Mean	Mean	Mean
Ploidy level (D, T)		1st year	2nd year	3rd year	Mean	leafiness	CP	CF
Maturity group (E, M, L)		of use	of use	of use		%	g kg <sup>-1</sup> DM	
Veja	D, M	10.8	3.78	4.51	6.36	38.4	131	260
Sodre	T, M	10.0	4.47	4.18	6.22	38.0	127	272
Zvilge	T, L	9.93	3.81	4.00	5.91	48.4	139	258
Respect	D, E	9.89	3.91	4.09	5.96	46.0	121	257
Merlinda	T, E	9.85	3.69	3.87	5.80	43.7	121	264
Recolta	D, M	10.3	3.71	4.09	6.03	62.3	139	244
Meltra	T, M	9.57	3.39	4.14	5.70	52.6	127	241
Veritas	D, L	10.5	3.52	4.37	6.13	52.7	141	255
Montagne	T, L	9.34	3.49	4.33	5.72	57.5	130	247
<i>LSD<sub>05</sub></i>					0.53	8.237	26.9	15.9

Ploidy level: D - diploid, T - tetraploid; Maturity group: E- early, M - medium-early, L - late.

The Lithuanian varieties Sodre and Veja produced the highest DM yield (6.22-6.36 t ha<sup>-1</sup>). The DM yield of Veja was significantly higher only than that of Merlinda, Meltra and Montagne. In all experimental years a trend was identified that tetraploid varieties produced a lower DM yield than diploid varieties.

The perennial ryegrass was under the influence of environmental factors across the years. Fisher's criterion ( $F_{\text{actual}}=1159.96 > F_{\text{theoretical } 0.1} = 6.23$ ) demonstrated significant yield differences. In the first year of use when the growing season was rainy and warm, ryegrass grew and yielded very well. Of the six foreign varieties tested the yield of the four varieties Respect, Merlinda, Meltra, Montagne was lower compared with all Lithuanian varieties. Only the varieties Recolta and Veritas yielded similarly to Sodre and Veja.

When comparing the varieties of the same maturity group but of different ploidy it is obvious that DM yield of diploid varieties was from 0.04 to 1.2 t ha<sup>-1</sup> greater than that of tetraploid varieties. The yield difference varied in relation to maturity, i.e. there was practically no difference between the early-maturing varieties (0.04 t ha<sup>-1</sup>), and between medium-early varieties the difference amounted to 0.73 t ha<sup>-1</sup> (and a similar yield difference (0.76 and 0.83 t ha<sup>-1</sup>) was obtained when comparing Lithuanian varieties). The highest yield difference (1.2 t ha<sup>-1</sup>) was obtained when comparing the late-maturing varieties.

In the second year of use, due to the shortage of rainfall (spring rainfall was 72% of the mean rate and there was no rainfall in August) and due to plant senescence processes, the herbage DM yield was low at 3.39-4.47 t ha<sup>-1</sup>; this was 5.53-7.02 t ha<sup>-1</sup> lower than in the first year of use. The Lithuanian variety Sodrė showed a high adaptability to adverse weather and local conditions. It produced a dry matter yield of 4.47 t ha<sup>-1</sup>.

Comparison of the varieties of the same maturity group but of different ploidy, showed that as in the first year of use the DM yield of foreign diploid varieties was 0.03-0.32 t ha<sup>-1</sup> higher than that of tetraploid ones, while an opposite situation was true for the Lithuanian varieties where the yield of diploid varieties was lower by 0.03-0.69 t ha<sup>-1</sup>.

In the third year of sward use the amount of rainfall and temperatures during the growing season were similar to long-term means. Although ryegrass plants thinned out after winter, the DM yield compared with that of the second year of use, increased by 0.18-0.85 t ha<sup>-1</sup>. The DM yield of grasses was almost the same (3.87-4.51 t ha<sup>-1</sup>) irrespective of the maturity group. The Lithuanian variety Veja was distinguished by higher productivity (4.51 t ha<sup>-1</sup>).

One of dry matter yield components is leafiness. Also, the quality of herbage is determined by leafiness, since leaves contain the largest amount of crude fibre compared with the other plant parts. In terms of leafiness, Lithuanian varieties were not superior to foreign ones. Late-maturing varieties were leafier. Averaged data suggest that the highest leafiness was identified for the Belgian variety Recolta. The lowest leafiness was recorded for the Lithuanian varieties Sodrė and Veja. Significant differences in leafiness were identified between the varieties. The coefficients of variation showed that the variety Recolta had the lowest variation of leafiness value over the whole experimental period.

The quality of perennial ryegrass varied in relation to weather conditions in the year assessment and varietal genetic differences. The content of crude protein was similar for all perennial ryegrass varieties tested. No significant differences were identified between the varieties. The variety Merlinda accumulated the lowest crude protein concentration (120.7 g kg<sup>-1</sup>), while Veritas accumulated the highest content.

The data analysis by ANOVA indicated a significant (54%) varietal effect on crude fibre concentration in the DM yield ( $F_{\text{actual}}=3.41 > F_{\text{theoretical } 0.5} = 2.59$ ). The content of crude fibre depended on the earliness of maturity and leafiness of perennial ryegrass. Significant differences in crude fibre content were identified when comparing early-maturing varieties Sodrė and Merlinda with medium-early or late-maturity foreign varieties. Leafier varieties accumulated less crude fibre. A strong negative correlation ( $r = -0.848^{**}$ ) was identified.

## Conclusions

Under western Lithuanian conditions diploid and early-maturing varieties were characterized by a higher productivity. All Belgian varieties were leafier compared with Lithuanian varieties. The varieties did not differ significantly in the crude protein content in the dry matter, and the lowest content of crude fibre was accumulated by the medium-early and late-maturity Belgian varieties.

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# Different management regimes of fodder galega (*Galega orientalis*) sward for organic farming

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## Abstract

Fodder galega can produce a high yield even without mineral fertilization and is suitable for soil conservation. Field trials were designed to identify the most suitable time for the last cut and to ascertain whether it is possible to alleviate the consequences of improper timing of the cut by using the sward under mixed management for forage and seed production. Due to the autumn cut of the aftermath, a significant reduction in herbage dry matter yield, metabolizable energy and digestible protein yield was obtained in the first and second cuts in the following year. A reduction in the seed yield occurred when the autumn aftermath was cut in August-September. When the autumn aftermath was taken annually for four years the productivity of the sward significantly declined when the cuts were taken during the August 30-October 15 period. The herbage yield produced over three cuts was significantly lower compared with the treatments cut twice. The herbage yield increase in the third cut did not compensate the losses that were incurred through the reduction in sward productivity. When the sward was used under mixed management for forage and seed production the negative effect of the third cut persisted; however, its effect was milder.

Keywords: *Galega orientalis*, forage production, last cut, seed yield

## Introduction

Fodder galega (*Galega orientalis*) is one of the least investigated perennial legume herbages. In Lithuania there has been increased interest in fodder galega recently. Farmers are looking for new legumes characterized by longevity and a high adaptability to growing conditions. Fodder galega is one of the longest-persisting plants in swards, and is notable for a high productivity without mineral fertilization (Virkajarvi and Varis, 1991; Adamovich, 2000). Fodder galega cultivation protects the soil from erosion and prevents the spread of weeds. Fodder galega can grow on a wide range of soils. It is important that ground water level is not high and the soils are not acidic (Raig *et al.*, 2001). In terms of its soil requirements, fodder galega is very similar to lucerne. Cut in the first week of June, fodder galega had similar contents of crude protein and fibre; however, the forage lagged behind lucerne in organic matter and feed unit yield (Moller *et al.*, 1997). Fodder galega has some drawbacks, such as very slow development in the sowing year and in the first year of use, and sensitivity to frequent cutting or grazing. Compared with other legumes, research into fodder galega cultivation issues is scarce. The objectives of the study were to identify the most suitable time for the last cut and to ascertain whether it is possible to alleviate the consequences of improper timing of the cut by using the sward under mixed management for forage and seed.

## Materials and methods

During 2000-2006 two field experiments were carried out on a sod gleyic loam soil (Epicalcari-Endohypogleic Cambisol) in the central part of Lithuania (55°23' N, 23° 51' E). The soil contained on average 2.63% of humus, 0.17N, 104 mg kg<sup>-1</sup> of P, 125 mg kg<sup>-1</sup> of K, and the was pH 7.0. Fodder galega (cv. 'Gale') was sown at a seed rate of 10 kg ha<sup>-1</sup> and at row spacing of 23 cm with a cover crop of spring barley. Field studies were continued for six



years in a fodder galega sward from the second to the seventh year of use. The experiment was designed as a randomized complete block with 4 replications and a plot size of 2.5 m × 8.0 m. When used for forage fodder galega was cut three times per season. The last cut was taken at a different time: on 15 August, 30 August, 15 September, 30 September, 15 October, 30 October. In the control treatment fodder galega was cut twice and the autumn aftermath was not cut. The first cut of galega was taken at the beginning of mass flowering (end of May/beginning of June). The second crop was taken at the end of first ten-day period of July, mass bud formation / beginning of flowering stage. When the sward was used under mixed management the use for forage was alternated with the use for seed. After-effects were monitored for the last two experimental years. In 2004 all galega plots were cut for forage (2 cuts, autumn aftermath was not cut), and in 2005 the galega plots were left for seed. Fodder galega did not receive any fertilization, and no chemicals were used.

## Results and discussion

In the year of trial establishment the swards of fodder galega contained few other plant species. Galega accounted for over 90% of the sward. With a delay in the third cut timing from August 15 to October 15, herbage dry matter yield increased from 0.92 t ha<sup>-1</sup> to 2.66-2.86 t ha<sup>-1</sup> (Table 1).

Table 1. Herbage yield and quality parameters of the last cut (third) of fodder galega, 2000.

Cutting date	Dry matter t ha <sup>-1</sup>	Pure galega t ha <sup>-1</sup>	Digestible protein kg ha <sup>-1</sup>	Metabolizable energy GJ ha <sup>-1</sup>	Crude protein g kg <sup>-1</sup>	Crude fibre g kg <sup>-1</sup>
15 August	1.05	0.92	149	11.36	221	320
30 August	1.93	1.80	202	18.94	168	355
15 September	2.86	2.75	252	27.15	137	350
30 September	2.60	2.52	268	25.87	160	342
15 October	2.66	2.59	264	25.80	154	317
30 October	2.32	2.21	165	21.06	121	418
LSD <sub>0.05</sub>	0.17	0.10	12	1.07	24	39

When the cut was taken at the end of October, due to the shedding of leaves herbage yield declined significantly and the quality of forage deteriorated. A significant reduction in digestible protein yield occurred, the content of crude fibre markedly increased in dry matter up to 418 g kg<sup>-1</sup>. In each experimental year, herbage yield of fodder galega's third cut depended considerably on the weather conditions, and when taken on October 15 it changed from 0.46 t to 2.86 t ha<sup>-1</sup>. This accounted for 8.2-33.0% of the annual yield. Metabolizable energy and digestible protein yield changed in proportion to the changes in fodder galega dry matter. Galega which is cut at the end of August or at the beginning of September starts to regrow again, as a result of which the plants' resources become depleted and are not capable of forming rhizomes or above-ground parts. It causes an essential decrease in the yield in the following year, and also inhibition of plant development (Raig *et al.*, 2001). Seed yield of the following year declined when autumn aftermath had been cut in August and September. When autumn aftermath had been cut annually for four successive years, the sward productivity significantly declined when the cuts were taken during the period 15 August-15 October. The herbage yield obtained from three cuts was significantly lower than that of two cuts. The extra yield of the third crop did not compensate for the losses resulting from sward productivity reduction. When the sward was used under mixed management for forage and seed the negative effect of the third cut persisted, but was weaker. When the sward had been used under mixed management for five years, galega accounted for 69-80% of the sward. In the swards that had been used only for forage the content of galega was lower, at

58-79%. Due to the autumn aftermath cutting, a significant reduction occurred in dry matter yield of herbage obtained during the first and second cut (Table 2).

Table 2. Herbage dry matter (t ha<sup>-1</sup>) and seed yield (kg ha<sup>-1</sup>) of galega.

Date of the 3rd cut	2000	2001	2002		2003	2004	2005	Mean of herbage DM yield t ha <sup>-1</sup>	Mean of seed yield kg ha <sup>-1</sup>
	3f	s	2f	3f	S	2f	s	2f-3f	s
Not cut	3.66	494	5.07		500	3.68	476	4.14	490
15 Aug	4.71	492	4.19	4.42	198	2.51	292	3.88	327
30 Aug	5.59	377	4.24	4.48	299	2.83	318	4.30	331
15 Sept	6.52	449	4.37	4.72	311	2.84	306	4.69	355
30 Sept	6.26	426	4.84	5.24	419	3.15	314	4.88	386
15 Oct	6.32	482	4.64	5.07	389	2.97	314	4.79	395
30 Oct	5.97	503	4.51	4.72	450	3.36	442	4.68	465
LSD <sub>0.05</sub>	0.67	63.7	0.45	0.46	70.2	0.19	46.0	0.48	60.8
	3f	2f	3f	2f	3f	2f	s	2f-3f	
Not cut	3.66	6.69		5.73		4.25	533	4.60	
15 Aug	4.71	4.95	6.36	3.46	3.84	2.58	328	3.80	
30 Aug	5.59	3.93	5.86	3.34	3.56	2.93	358	3.93	
15 Sept	6.52	4.86	6.96	3.93	4.32	3.04	349	4.59	
30 Sept	6.26	5.46	7.37	4.33	4.99	3.26	386	4.86	
15 Oct	6.32	5.70	7.43	4.95	5.35	3.38	496	5.03	
30 Oct	5.97	6.07	7.97	5.01	5.18	4.17	534	5.31	
LSD <sub>0.05</sub>	0.67	0.34	0.42	0.53	0.51	0.46	53.5	0.49	

f- sward used for forage; 2f-2 cuts; 3f- 3cuts; s- sward used for seed.

The greatest damage to the sward is done when autumn aftermath is cut during the period from August 15 to September 30. In Estonia the later cutting at the end of growing period in October had a favourable effect on galega growth and the spring yield of the following year (Raig *et al.*, 2001). However, the sward there was cut only twice and the effect of the last cut was compared when the cut was taken on September 12 and October 23. Autumn aftermath cutting (15 August-15 October) significantly reduced the number of generative shoots, stem height and thickness, and the number of pods, but had no effect on seed quality.

## Conclusions

Fodder galega is very sensitive to autumn aftermath cutting. It is more expedient to use young galega swards for seed or cut only twice per season, and if autumn aftermath is needed, it should be taken in the middle of October, towards the end of the growing season, so that it does not re-grow. When galega is used under mixed management for forage and seed, the negative effects of autumn aftermath cutting are alleviated or eliminated more rapidly.

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## Production and morphological traits of tall oatgrass breeding families

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### Abstract

Tall oatgrass (*Arrhenatherum elatius*) is widespread in Serbia in grasslands of anthropogenic origin, particularly at lower elevations but also to 1200 m a.s.l. As a secondary grass species, tall oatgrass has lower fodder quality in the dry matter, but high and stable yield in difficult ecological conditions classifies it among perennial grasses suitable for drought and poor soil, and in Serbia tall oatgrass is a component of grass-clover mixtures for livestock feed production. In this work seven breeding families that originated from polycross of tall oatgrass plants and two synthetics were examined in comparison with a standard cultivar, K-12. Morphological traits, components of dry matter production and quality (plant height, leaf size and number, panicle length, leaf-stem ratio, number of tillers), time of tillering, dry matter yield and regeneration were investigated in a 3-year plot trial. Based on productivity and the agronomically most important traits, all polycross families were considered suitable for further breeding, while synthetics could be used as new synthetic cultivar.

Keywords: tall oatgrass, dry matter yield, morphological traits, breeding, polycross, synthetics

### Introduction

Tall oatgrass (*Arrhenatherum elatius* (L.) P. Beauv. ex J. Presl & C. Presl.) is an indicator species of the *Arrhenatheretum elatioris* association, considered to be among the highest quality natural lowland meadows in Europe. In Serbia, this association is of anthropogenic origin and is mostly in lower regions (Jovanović-Dunjić, 1983), but tall oatgrass can also be found to 1200 m a.s.l. Tall oatgrass is a perennial forage grass that belongs to the sub Central-European floristic element and has a distribution throughout almost the entire area of Serbia, including low and hilly-mountainous regions. In mixture with other grasses it is persistent over time due its broad ecological amplitude and resistance to cold and drought. It grows best on fertile and well-drained soils, but also on other soil types (Swedrzynski and Kozłowski, 1998). It has high and stable forage yield and therefore is one of the significant forage grasses for mixtures especially in conditions of drought and poor quality soils in Serbia. Tall oatgrass shows somewhat lower forage quality than other forage grasses. However, according to Čolić *et. al* (1986) this trait can be significantly improved by changing the cutting frequency and increasing the proportion of leaves in total dry matter (DM). It can also be improved by breeding for quality and increased crude protein (CP) content while maintaining a high forage yield. A final step in the breeding process is to test production and other important traits with standard cultivars, which was the aim of this experiment.

### Materials and methods

The trial was established on an experimental field of the Institute for Forage Crops, Kruševac, at 142 m a.s.l. Seven F1 breeding families (P 7/18, P 8/6, P 11/7, P 11/17, P 11/21, K 13/18 and P 19/16) which originated from polycross of chosen tall oatgrass plants, and two synthetic breeding populations (Synth. 1 and Synth. 2), were included in the experiment. All families and populations were examined in comparison with the widely used standard cultivar K-12. Breeding families and synthetics originated from landraces that were collected on farms in

Central Serbia from localities Pasjak (P) and Kobilje (K). The trial was sown in 2m<sup>2</sup> plots with three replications in spring 2004. Morphological traits, components of dry matter production and quality in the first cut (plant height, number of generative and vegetative tillers, leaf-stem ratio, leaf width, length and number, panicle length) and dry matter yield were investigated over a 3-year period. Dry matter yield (DMY) was determined in two and four cuts, depending on the investigation year and drought effect, and analysed by ANOVA. Number of tillers was calculated per m<sup>2</sup>. Time of tillering (days from April 1st) and regeneration height 14 days after the first cut were also recorded. Leaf : stem ratio was analysed on a dry matter basis and also presented as a percentage.

## Results and discussion

Genotypes (plants) included in polycross were chosen according DMY and leaf size components. It is expected that breeding tall oatgrass families have improved DMY and leafiness, as a prerequisite of good herbage quality. There is significant variability between families for DMY (Table 1). The maximum average annual DMY (14.53 t ha<sup>-1</sup>) was obtained in synthetic breeding population 2. That yield was exactly 1 t ha<sup>-1</sup> greater than the DMY of the standard cultivar. Breeding families showed mean annual DMY of *ca.* 13 t ha<sup>-1</sup>, except P 7/18 (11.67 t ha<sup>-1</sup>). The best yielding breeding family was K 13/18 with 13.4 t ha<sup>-1</sup> of DM. In the first investigation year the most productive was synthetic breeding population 1 with 13.8 t ha<sup>-1</sup>, and almost equal DMY in each cut (7.0 and 6.8 t ha<sup>-1</sup> respectively). In the second year, with higher annual DMY (> 18 t ha<sup>-1</sup>) the first cut gave more than 50% of annual DMY. At the first cut the most productive were synthetic breeding population 1 and breeding family K 13/18, with > 11 t ha<sup>-1</sup>. Synthetic breeding population 2 in the second year gave a total DMY of > 22 t ha<sup>-1</sup>. In the third year DMY was depressed by drought and reached only 7 to 9 t ha<sup>-1</sup>. The exception was synthetic breeding population 2; annual DMY was 12.2 t ha<sup>-1</sup>, and it probably had better drought tolerance than other families.

Table 1. Dry matter yield (t ha<sup>-1</sup>) of tall oatgrass families and synthetic breeding populations.

Families	First year			Second year					Third year			Average annual DMY
	I cut	II cut	Total	I cut	II cut	III cut	IV cut	Total	I cut	II cut	Total	
P7/18	5.6	5.1	10.7	9.9	3.9	2.2	0.9	16.9	5.4	2.0	7.4	11.67
P8/6	6.6	5.4	12.0	10.5	3.6	2.7	0.8	17.6	5.9	2.4	8.3	12.63
P11/7	6.8	5.8	12.6	9.8	5.3	2.7	1.1	18.9	5.5	2.3	7.8	13.10
P11/17	6.9	5.4	12.3	9.4	5.2	3.0	1.3	18.9	5.7	2.7	8.4	13.20
P11/21	7.3	5.6	12.9	10.4	4.0	2.7	0.9	18.0	5.3	2.3	7.6	12.83
K13/18	7.3	5.8	13.1	11.2	4.0	2.6	1.0	18.8	6.0	2.3	8.3	13.40
P19/16	7.4	5.6	13.0	10.1	4.1	2.9	1.2	18.3	5.9	2.6	8.5	13.27
Synth. 1	7.0	6.8	13.8	11.6	4.3	2.5	0.9	19.3	6.4	2.4	8.8	13.97
Synth. 2	3.3	5.8	9.1	9.5	7.3	3.6	1.9	22.3	8.2	4.0	12.2	14.53
K-12	7.7	5.9	13.6	10.3	3.8	2.1	1.2	17.4	7.1	2.5	9.6	13.53
Lsd 005			1.92					2.92			2.21	1.37
001			2.63					3.99			3.02	1.87

Time of tillering is ranked for all populations and families in 4 days in early May (Table 2). For morphological traits, variations of mean values differed significantly between populations (CV between 7.57 and 20.86%). At the first cut, average height of plants was *ca.* 101 cm, while regrowths were 11-16.4 cm. Population P 8/6 had the highest plants (113.3 cm) at first cut, while its regeneration was the lowest (11cm). Breeding families P 11/7 and P 11/17 had the most leaves per plant as well as largest size of leaf sheath. These morphological traits affect forage quality by increasing proportion of leaves in total dry matter and improving

protein content. All populations and families showed improved leafiness in comparison with K-12. Ševčíkova and Šramek (2002) reported somewhat lower mean values of tall oatgrass morphological traits in investigations of a European collection. Average panicle length, an important trait for seed yield, was 17.1 cm, but could reach 21.5cm (Kozłowski *et al.*, 1998).

Table 2. Investigated traits of tall oatgrass families and synthetic breeding populations (3-year mean values).

Traits	Time of tillering	Plant height (cm)	Regeneration (cm)	No. of vegetat. tillers	No. of generat. tillers	Leaf: stem ratio (%)	Leaf width (mm)	Leaf length (cm)	No. of leaves per tiller	Panicle length (cm)
Families										
P7/18	33.5	105.0	16.2	1232	1120	25.9:74.1	10.3	22.3	3.7	18.7
P8/6	36.5	113.3	11.0	1040	1872	23.1:76.9	9.0	19.3	4.7	19.0
P11/7	34.5	105.7	15.3	560	3520	20.4:79.6	8.3	26.0	4.7	16.7
P11/17	37.5	95.3	13.6	1536	1568	29.3:70.7	10.0	27.3	4.7	17.7
P11/21	37.5	93.0	16.4	736	1856	20.7:79.3	6.7	20.7	4.3	15.7
K13/18	34.5	105.0	16.4	976	1600	21.8:78.2	7.3	21.7	3.7	19.3
P19/16	34.5	108.3	12.7	816	992	22.8:77.2	9.3	27.7	3.3	14.3
Synth. 1	34.5	89.7	12.0	896	1088	24.8:75.2	7.7	22.3	4.0	12.7
Synth. 2	34.5	105.0	15.7	1200	1440	23.2:76.8	11.0	22.7	3.3	18.0
K-12	34.5	95.0	12.7	576	1600	18.4:81.6	5.3	20.3	3.7	19.0
Average	35.2	101.5	14.2	956.8	1666	23.1:79.6	8.5	23.0	4.0	17.1
CV (%)	4.03	7.57	14.29	32.16	43.26		20.86	12.83	13.93	13.08

The largest number of tillers per surface area was found in breeding family P 11/7 (4080), but the ratio between generative and vegetative shoots, which can improve forage quality, was more suitable in families P 11/17 and P 7/18 (almost 50:50). This ratio was also good also in synthetic 2 and family P 8/6. Variability among families in the number of tillers was also highest for generative tillers (43.3%). In other research, Kozłowski *et al.* (1998) found lower numbers of generative tillers, 386-683 m<sup>-2</sup>. The best leaf : stem ratio in the total herbage was present in families which also had the best leafiness (P 11/17 and P 7/18) and in synthetic 1. All genotypes had better leaf content in the total herbage than cultivar K-12.

## Conclusions

Most breeding families and synthetic breeding populations showed improved morphological traits and production in comparison with the cultivar K-12. The best yield results were obtained for synthetic population 2 and breeding family K 13/18, while the highest values of morphological traits were recorded for breeding populations P11/17, P7/18 and synthetic population 2. Based on results on productivity and for the agronomically most important traits, all polycross families are suitable for further breeding, while the synthetic population 2 could be used as a new synthetic cultivar.

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# Manipulating pasture grass growth by nitrogen fertilization

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## Abstract

In two field trials on sown and permanent grassland the possibilities of influencing grass growth of pastures via the seasonal distribution of mineral nitrogen (N) fertilization was studied over four years in the region Bern-Solothurn. In the four treatments the annual fertilizer application of 150 kg N as ammonium nitrate was distributed as follows: evenly distributed over the whole growing season or higher doses in spring, summer or autumn. Three of the four experimental years had pronounced dry periods during summer which lead to a considerable decrease of dry matter (DM) growth and annual yields of only 10-11 t ha<sup>-1</sup>. In spite of this, the seasonal distribution of grass growth could be substantially varied through the seasonal distribution of N fertilization. It was possible to shift approximately 10% of the total annual yield from spring to late summer and autumn. The seasonal distribution of the N fertilizer did not significantly influence the total annual yield. On average, the N fertilization increased the total annual pasture yield by 27%, as compared to the unfertilized control treatment. On average over all experimental sites, years and treatments the N effect was 16.3 kg DM per kg N. The results imply that a tactical distribution of N fertilization could help to harmonize pasture growth and feed requirements. Particularly on pastures with less than 25% clover the tactical distribution of N fertilization can help to simplify pasture management through a more even growth rate. This is especially true for the higher doses in summer.

Keywords: Nitrogen application pattern, grass growth, herbage mass, extended grazing season

## Introduction

Mineral nitrogen (N) fertilizer use is quite severely restricted in Switzerland by policy measures. On full grazing farms the use of mineral N should therefore not primarily be used to maximize yields, but rather to harmonize feed growth with feed requirements. Studies in England and Ireland have shown that the course of the grass growth curve can be influenced by a tactical distribution of N fertilization (Binnie *et al.* 2000; Hennessy *et al.* 2004). Nitrogen fertilization during winter can promote grass growth in spring, while N fertilization in late summer or in autumn can extend the grazing period by up to one month. Two questions are especially relevant: (1) What flexibility can be achieved in the lowlands of Switzerland through a tactical distribution of N fertilization with respect to the desired harmonization of roughage availability and feed need? (2) What yield effect is achieved per unit N if the N fertilization pattern is varied?

## Materials and methods

A sown grass white clover meadow (Swiss standard mixture SM 440) and an intensive permanent pasture in the region Bern-Solothurn were selected for the experiment. On both sites the main grasses were *Lolium perenne* and *Poa pratensis*. Both sites were on full grazing farms with continuous grazing system (Table 1). Five treatments were compared, four with differing distribution of 150 kg N ha<sup>-1</sup> plus a non-fertilized control: 1) control, 2) seven equal doses of 10-25 kg N ha<sup>-1</sup>, 3) spring-accentuated with five doses of 30 kg N ha<sup>-1</sup> in spring and summer, 4) summer-accentuated with 5 doses of 30 kg N ha<sup>-1</sup> from late spring to early autumn and 5) autumn-accentuated with five times 30 kg N ha<sup>-1</sup> in March, May, June and September

(two times). A grass growth curve according to the method of Corall and Fenlon (1978; slightly adapted) was established for each treatment on each of the two sites over the four years of the project duration (2003-2006; Figure 1). A new site of 300 m<sup>2</sup> was fenced in each year for the growth curve. Three repetitions were used for each treatment and cutting regime. Three of the four years were characterized by a pronounced dryness period in summer. The precipitation during June, July and August was 40%, 57% and 66% below the long-term average for 2003, 2005 and 2005, respectively. In 2003 the average summer temperature (July to August) was 5 °C above the long-term average.

## Results and discussion

The comparison of the different fertilization treatments shows that total yield was hardly affected by the distribution of mineral N fertilizer (Table 1). This is in agreement with the results of previous experiments, e.g. Caputa and Schechtner (1970), Morrison (1980), Thomet and Brühlmann (1987) and Hennessy *et al.* (2004).

Table. 1. Total annual dry matter yield (kg DM ha<sup>-1</sup>) and nitrogen effect (kg DM kg<sup>-1</sup> N) of the four treatments with variable seasonal distribution of 150 kg N ha<sup>-1</sup> and the unfertilized control treatment for the two experimental sites Bremgarten and Hessigkofen.

	Bremgarten 2003 & 2004		Hessigkofen 2003 & 2004		Bremgarten & Hessigkofen 2005 & 2006	
% clover	25-37		3-11		3-19	
Fertilizer treatment	Yield	N effect	yield	N effect	Yield	N effect
Unfertilized	12410 <sup>a</sup>	-	8850 <sup>a</sup>	-	7300 <sup>a</sup>	-
equal distribution	13560 <sup>ab</sup>	7.7	10930 <sup>b</sup>	13.9	10760 <sup>b</sup>	23.1
Spring accentuated	13250 <sup>ab</sup>	5.6	10890 <sup>b</sup>	13.6	10280 <sup>b</sup>	19.9
Summer accentuated	13710 <sup>b</sup>	8.7	11290 <sup>b</sup>	16.3	10400 <sup>b</sup>	20.7
Autumn accentuated	13430 <sup>ab</sup>	6.8	10640 <sup>b</sup>	11.9	9920 <sup>b</sup>	17.5

<sup>a, b</sup> different letters mean, that treatments are significant different ( $P < 0.05$ );

General Linear Model GLM and Tukey-Kramer test.

Summer dryness, yields were clearly below the 12500 kg ha<sup>-1</sup> expected in the lowlands of Switzerland in 3 of the 4 years. Hessigkofen was less productive than Bremgarten, especially in 2003 and 2004. The N-response was much lower at Bremgarten in the first two years after sowing, when the clover content was still above 25%. The seasonal growth curve was mainly influenced by weather conditions. In summer rainfall was the limiting factor, in spring and autumn temperature. Nevertheless, sward growth could be influenced considerably through N fertilization. A summer pronounced N distribution helped smoothen the growth curve (Figure 1). As compared to the spring-accentuated treatment, 1,160 kg DM ha<sup>-1</sup> or approximately 10% of the annual yield was shifted from spring to autumn. For the period with decreasing growth (August to November) the roughage availability was thus increased by 45%. This would be equivalent to 63 cattle unit grazing days or an extension of the grazing period by approximately three weeks. The autumn accentuated treatment (not shown in Figure 1) did not achieve an additional benefit but the lowest annual yields.

The seasonal distribution of N fertilization also influenced the clover content of the swards. Irrespective of the treatment, the highest content of white clover was reached in July. The



highest proportion of clover was achieved by summer-accentuated N fertilization. Compared to the spring-accentuated treatment the proportion of clover in the June-August total yield was *ca.*10% higher. This will also increase roughage consumption and the milk production potential. In 2003 and 2004 the NO<sub>3</sub> content of the roughage was analysed in summer and autumn; it was increased by summer- and autumn-accentuated N fertilization. Compared to the even-N distribution the increase was 1,000-1,500 mg NO<sub>3</sub><sup>-</sup> kg<sup>-1</sup> DM. Only for two weeks at the beginning of October 2003 a level that might be considered critical from the animal health point of view was reached (of 5000-5500 mg).

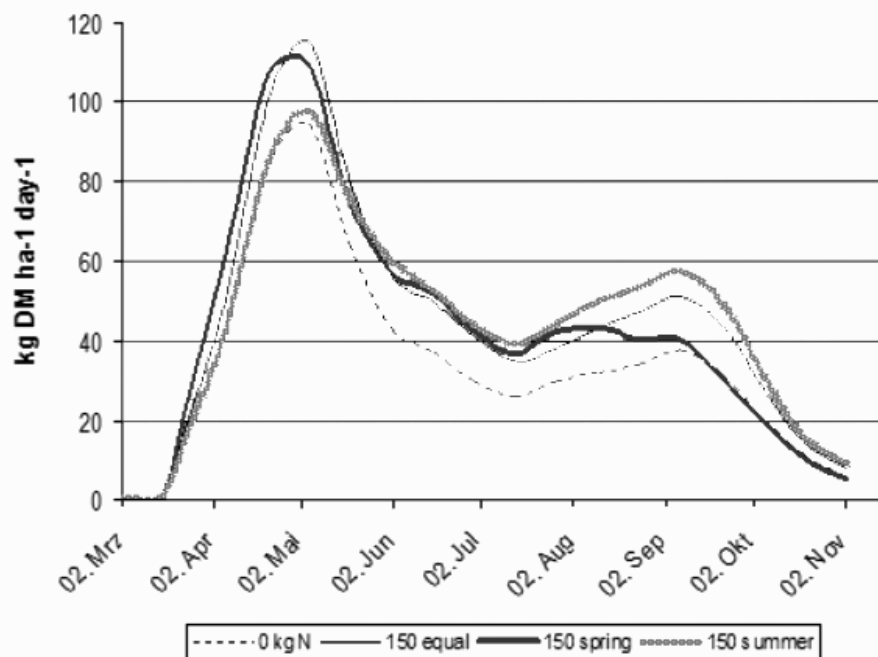


Figure 1. Influence of the seasonal distribution pattern of mineral N fertilizer on the growth curve (average over two sites and four experimental years).

## Conclusions

The experiments showed that it is possible without effect on total yield to shift approximately 10% of the annual yield from spring to autumn with the seasonal distribution of N fertilization. Thus the grazing period can be extended in autumn. It can be concluded that summer-accentuated N fertilization is most promising for full grazing dairy production in the lowlands of Switzerland because it allows a more constant stocking rate, an extension of the grazing period in autumn, a high N efficiency and a higher clover content of the swards.

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# Influence of site conditions on interspecific interactions and yield of grass-legume mixtures

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## Abstract

Composition and yield of mixtures including fast- and slow-growing grasses (*Lolium perenne* and *Dactylis glomerata*) and fast- and slow-growing legumes (*Trifolium pratense* and *Trifolium repens*) at two sowing densities were explored over three years. Results are presented from two sites in southern Germany with different environmental conditions but similar management. The investigation is part of COST action 852 Working Group 2. Due to interspecific interactions the botanical composition of the mixtures altered. The proportion of legumes declined, *D. glomerata* became more and more competitive, and *T. pratense* showed the lowest persistence. These results were obtained at both sites, only the rate of change being different. At four cuts per year the mixtures achieved yields up to 17 Mg DM ha<sup>-1</sup> at site I and up to 15 Mg DM ha<sup>-1</sup> at site II. The fast-growing species produced the highest yields in the first and second year, while *D. glomerata* was more productive afterwards.

Keywords: interspecific interaction, grass-legume mixtures, forage yield, sowing density, persistence

## Introduction

Intensive grassland systems are widespread and cover large areas. They are very important for fodder production and are increasingly used for biogas production. High-yielding grass-legume mixtures with appropriate chemical and physical forage characteristics have to be established for these purposes. To achieve a satisfactory botanical composition, knowledge of the interactions between species during different phases of sward establishment is necessary. The modification of competition between different sown species and weeds in plant communities by various environmental conditions also has to be kept in mind.

Within Working Group 2 of COST action 852, mixtures of slow- and fast-growing grasses and legumes were tested at twenty-eight sites over a large environmental gradient in Europe (Kirwan *et al.*, 2007). Dry matter yield and botanical composition from two sites with different environmental conditions in south-west Germany are presented here.

## Materials and methods

Thirty plots per site were established in a simplex design (Cornell, 2002) with two grass (*Lolium perenne* 'Lacerta', *Dactylis glomerata* 'Accord') and two legume species (*Trifolium pratense* 'Merviot' and *T. repens* 'Milo'). *L. perenne* and *T. pratense* are fast-establishing species while *D. glomerata* and *T. repens* are known as slow-establishing but more persistent. We used eleven mixtures of the four species and four monocultures. Four mixtures were dominated by one species (70% of the seed rate in monoculture and 10% of each of the other three species), six mixtures were dominated by a pair of species (40% of the two species, 10% of the other two), and one mixture had equal proportions (25% of each species). The basic seed rate (monoculture) was 35 kg ha<sup>-1</sup> *L. perenne*, 25 kg ha<sup>-1</sup> *D. glomerata*, 12 kg ha<sup>-1</sup> *T. pratense*, and 35 kg ha<sup>-1</sup> *T. repens*. The monocultures and mixtures were sown at two seed



rates (normal seed rate and 40% reduced seed rate). As there were small differences in the results for the two seed rates only the mean values are shown here.

Site I (altitude 460 m a.s.l., annual rainfall 693 mm, mean daily temperature 8.1 °C) is characterized by good soil conditions (arable land, silty loam), while site II (altitude 700 m a.s.l., annual rainfall 834 mm, mean daily temperature 6.6 °C) is a rendzina with shallow soil depth. Fertilization (150 kg N ha<sup>-1</sup> y<sup>-1</sup>) and cutting frequency (four cuts per year) are typical of intensively used grassland in this region.

In this paper the results of the three experimental years following the sowing year are presented (site I: sowing year 2003, results of 2004-2006, site II: sowing year 2004, results of 2005-2007).

The simplex design without replications is targeted at modelling mixture effects; therefore the data could not be subjected to a conventional statistical analysis.

## Results and discussion

Between the two sites there were marked differences in the development of yield proportions of the four species under investigation (Figure 1). At site I the proportion of *L. perenne* generally decreased, even if there was an increase in some plots in 2005. *L. perenne* is a species with fast germination which is initially very competitive but usually has no marked persistence. At site II it was damaged in winter 2005/2006 due to long-lasting snow cover but could regenerate during the following year. Yield proportions corresponded to the percentage of *L. perenne* in the seed mixture.

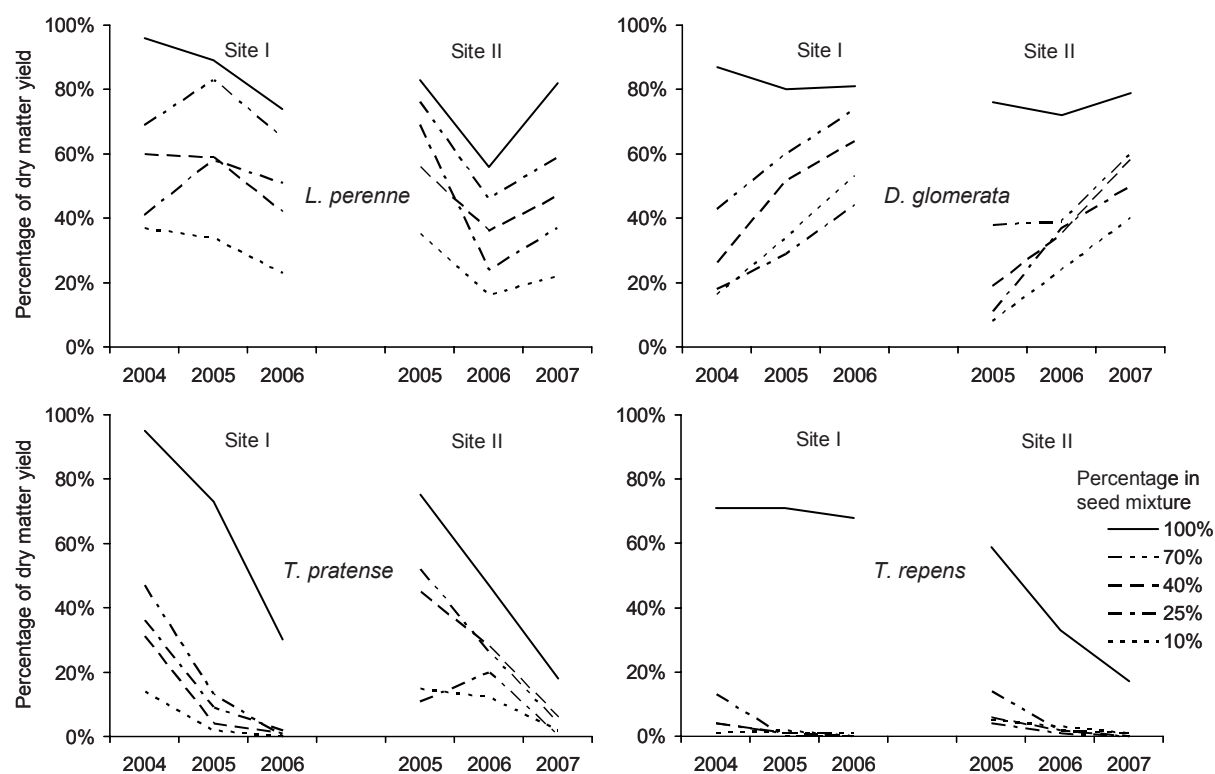


Figure 1. Development of yield proportion of the four species depending on their percentage in the seed mixture. The values shown are means over four harvests per year.

*Dactylis glomerata* showed a different response. In the first year, as a result of slow germination, it only had a major yield percentage in monocultures without interspecific competition (besides that of weeds). Over the years the proportions increased continually, at

site II nearly independently of the percentage in the seed mixture. *Dactylis glomerata* was able to use the space which was not longer occupied by *L. perenne* and particularly by *T. pratense*. *Trifolium pratense* grew well in the sowing year, but the ratio declined rapidly afterwards. In monocultures and mixtures with high proportions of *T. pratense*, weeds could establish and, especially in mixtures, attained high proportions. At both sites *T. repens* had no possibility to establish in the mixtures. Contrary to expectation, it did not increase its yield percentage in the years after sowing. The management and fertilizing regime probably encouraged the grasses too much.

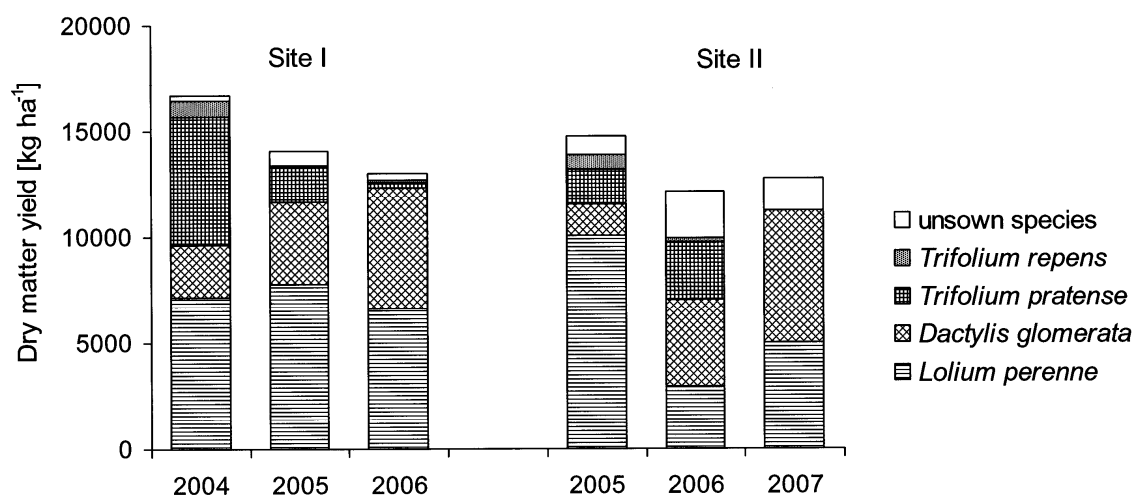


Figure 2. Dry matter yield (sum of four harvests) of the swards from the seed mixture with equal proportions of the four species (25:25:25:25%).

Dry matter yield was highest in the first year after sowing at both sites (Figure 2). *Lolium perenne* and *T. pratense* are responsible for this very high value. In the following years the total yield declined but the proportion of *D. glomerata* rose (Figure 2). At site II unsown species (mostly *Poa trivialis*) also had the possibility to establish. Due to the decline of *L. perenne* in winter 2005/2006 there was no sown species that could spread quickly enough to compete with the weeds. It is assumed that in subsequent years yield will be more constant because the more and more dominant species *D. glomerata* is not very susceptible to unfavourable weather conditions or other injuries. The yields of the mixtures always exceeded those of the monocultures.

## Conclusions

Mixing fast- and slow-growing grasses and legumes can enhance productivity in the first years after sowing. Under favourable site conditions the replacement of species succeeded without establishment of weeds, but the risk of an undesirable development is still high. The results show that it is very important to establish species like *D. glomerata* which are competitive in the years after sowing and have the ability to build stable swards by using space released by other species.

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## Leguminous species in ass. *Agrostetum vulgaris* and ass. *Festucetum valesiaca* on Stara Planina mountain

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### Abstract

In the hilly-mountainous region of Serbia, meadows and pastures provide the grazing and fodder that forms the basis of production for large and small ruminants. The objective of the work reported in this paper is to determine the share of useful leguminous plants and other species and weeds in three locations of ass. *Agrostetum vulgaris* and at four locations of ass. *Festucetum valesiaca* and, on the basis of the data obtained, to determine their value in ruminant nutrition. Based on the obtained data, the presence of useful species of leguminous plants varied in the investigated associations as well as the locations within same association. The highest presence of useful legumes was found in ass. *Agrostetum vulgaris* at location III (39.13%), and in ass. *Festucetum valesiaca* at location IV (42.86%). This is more than half of all useful species on grassland. The following useful legumes had the highest presence in all the analysed associations: *Trifolium ochroleucum*, *T. incarnatum*, *T. incarnatum*, *Lathyrus hirsutus*, *T. pratense*, *T. repens*, *T. campestre*, *T. montanum*, *Lotus corniculatus*, *Lathyrus montanus*, *Vicia grandiflora* and others. The results obtained provide a basis for improvement measures relating to the quality of grassland by applying melioration measures.

Keywords: floristic composition, useful legumes, ass. *Agrostetum vulgaris*, ass. *Festucetum valesiaca*

### Introduction

The mountain of Stara Planina, according to its geographical, climatic, orographic and other characteristics, represents a typical hilly-mountainous region of Serbia with livestock production as the main economic activity. However, over a long period of time the grassland meadows and pastures have begun to show signs of abandonment due to the decrease in the number of head of livestock and depopulation of villages. As a consequence, production in this hilly-mountainous region, as in the rest of Serbia, has almost ceased to exist. There is now a need to revitalize the economy, and the revitalization includes the analysis of grasslands and pastures as a prerequisite for new exploitation systems and market orientation in livestock production. Klapp (1986) noted that until the 1930s, and even today, only species from the family of grasses (*Poaceae*) and legumes (*Fabaceae*) were considered to be useful and desirable, and other species were considered as worthless and even harmful. The main criteria in the evaluation of whether a meadow plant is weed or not are the following: whether it is consumed by livestock, its nutritious value and digestibility, whether it is poisonous and to what degree, and whether it is suitable for use by domestic animals in relation to its morphological structure. In the investigations by Tomić *et al.* (2003a) on 12 locations on Stara Planina Mountain it was determined that the presence of legumes varied from 0-50%. The objective of this research was to determine the presence of leguminous plants on natural and artificial grasslands, as the most important group of plants not only in regard to the quality of grassland, but also as an indicator of soil characteristics and more appropriate use of agro-technical measures, especially NPK and PK fertilizers.

## Materials and methods

At the end of July, the year's yield of grasslands on Stara Planina Mountain was determined at on seven locations at an altitude of 650-850 m. Samples were taken on two occasions from an area of 1m<sup>2</sup> in order to determine the floristic composition of associations. Based on the weight share the percentage of individual species on the grassland was calculated and qualitative categorization of species according to Kojić (1990, 2001) was carried out.

## Results and discussion

Legumes represent an important plant group in the floristic composition of natural grasslands. They have an extremely important role in improving soil with nitrogen due to the symbiotic relation with bacteria which bond nitrogen from the air. Leguminous plants appear often in cycles on grasslands. When the share of legumes is declining, the share of grasses and other plants is increasing. The presence of legumes is often presented in average in all previous research. Könekamp (1965) reported that the legume share for German meadows situated in valleys was 10%, and on mountain grasslands 20%. On seven locations on Stara Planina Mountain, Tomić *et al.* (2003b) determined the presence of leguminous plants was in the range of 0-42.4%. Table 1 presents the presence of useful legumes, useful grasses and other useful species, and also weeds, as well as the total presence of useful species for both ass. *Agrostietum vulgaris* and ass. *Festucetum vallesiaceae* on seven locations. The floristic composition of plant associations on three locations (650-690 m a.s.l.) indicates typical ass. *Agrostietum vulgaris*, whereas the presence of legumes differs between the highest as determined on location I, at 39.13%, also very high on location III, at 34.62%, and the lowest on location II, at 19.23%. The presence of useful species was found to be in the same order.

Table 1. The percentage of useful legumes and other plant groups in ass. *Agrostietum velgaris* (3 locations) and ass. *Festucetum vallesiaceae* (4 locations).

Association <i>Agrostietum vulgaris</i>					
Location	Useful legumes	Useful grasses	Other useful plants	Weeds	Total useful
Location I (Gulenovci 650 m)	39.13	34.78	4.35	21.74	78.26
Location II (Gulenovci 680 m)	19.23	30.77	3.85	46.15	53.85
Location III (Gulenovci 690 m)	34.62	26.92	-	38.46	61.54
Association <i>Festucetum vallesiaceae</i>					
	Useful legumes	Useful grasses	Other useful plants	Weeds	Total useful
Location I (Mojinci 750m)	18.75	18.75	15.63	46.87	53.13
Location II (Mojinci 810m)	31.82	9.09	18.18	40.91	59.09
Location III (Mojinci 830m)	23.67	13.18	15.78	47.37	52.63
Location IV (Mojinci 850m)	42.86	22.86	17.14	17.14	82.86

The presence of weed species was found to be in inverse proportion. Nine species of legumes were found on each of locations I and III, and on both these locations the most abundant species was *Trifolium ochroleucum*. At location II 5 species were found, and the most

abundant was *Medicago lupulina*. According to the floristic composition of meadow associations on Sjenica-Pester highlands, determined by Mrfat-Vukelić *et al.* (2003), the presence of certain useful species (according to their weight share) in ass. *Agrostietum vulgaris* varied from 56.0-98.1%, whereas weeds were present in amounts between 1.9% and 44.0%. On four locations (at 750-850 m a.s.l.) ass. *Festucetum vallesiacae* was found with the presence of legumes ranging from 18.75% at location I, up to 42.86% at location IV. At location II legumes were present with 31.82%, and on location III with 23.67%. The most abundant species in all locations were: *Trifolium campestre*, *Medicago lupulina*, *Lotus corniculatus* and *Trifolium ochroleucum*. The total proportions of useful species in these associations varied from 52.63% to 82.86%. Weed species were present on the first three locations in very high proportions, as much as 40% or more of the sward.

## Conclusions

The floristic composition of plant associations indicates typical ass. *Agrostietum vulgaris*, whereas the presence of legumes differs from the highest proportions at location I (3.13%), and also very high on location III (34.62%), to the lowest on location II (19.23%). Ass. *Festucetum vallesiacae* were found with the presence of legumes from 18.75% on location I, up to 42.86% on location IV. For the next year melioration measures are recommended, with fertilization using optimal doses of NPK fertilizer in order to eliminate undesired species from associations and develop the desired species from either *Poaceae* or *Fabaceae* families.

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# Radiation use and uptake of mineral N in leys under climatic change

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## Abstract

A mechanistically based model was used for assessing the effect of climatic change on ley production. The model was tested against field trials in southern Sweden. The calibration to above-ground biomass and N concentration resulted in the radiation-use efficiency decreasing with age and after cutting, and increasing with nitrogen fertilization. The N-uptake efficiency of soil mineral N varied in a similar way to the growth period. The climate change assessments suggest that the growth potential might increase considerably by the end of the current century and that N-fertilization rates need to be increased to fully utilize this potential.

Keywords: growth model, calibration, field experiment, scenario

## Introduction

In estimating the impact of climatic change on ley production, it is essential that the plant production models used have been calibrated for a wide range of environmental conditions. For this purpose a large-scale experimental series, conducted by the Regional Experimental Service of Sweden during 1985-88, was used to assess the production of climate change scenarios for the next 100 years.

The aims were to estimate how site conditions, cutting and fertilization influence utilization of environmental resources (radiation and nitrogen) by grass/clover leys, and to use the calibration for 1985-88 (Eckersten *et al.*, 2007a) to predict the effects of climate change for about 100 years later (Eckersten *et al.*, 2007b).

## Material and methods

To simulate growth as function of climate it was set proportional to solar radiation absorbed by the canopy, complemented by the partitioning of assimilates between shoot and root and regrowth after cutting, following principles for carbon assimilation and allocation. When available N was non-limiting, N uptake was regulated by the maximum plant N needed for growth, and by the radiation conditions in terms of day length and plant biomass. N uptake under N-limiting conditions was regulated by the soil mineral N and root performance, here simulated as the daily uptake being proportional to mineral N in the root zone. Water conditions influence both plant growth and soil processes (see further Eckersten *et al.*, 2007a). Radiation use, root/shoot allocation, regrowth, the maximum N concentration and N uptake efficiency were calibrated against field data of pure grasses (*Phleum pratense* L. + *Festuca pratensis* Huds.), pure *Trifolium pratense* L., and a mixture of these, grown in 1985-88 at three N levels (0, 14 and 20 g N m<sup>-2</sup>) at five locations close to Uppsala, Jönköping, Skara, Örebro and Halmstad, respectively. A two-year life cycle was sampled for above-ground dry matter and N in two growth periods, during the first and the second year. The climate change assessments were based on climate change scenarios from the Rossby Centre (SMHI; dated October 2006) derived from emission scenarios A2 and for the period 2071-2100. Effects of

climatic change with and without CO<sub>2</sub> increase were simulated assuming unchanged plant N status (see further Eckersten *et al.*, 2007b).

## Results

The calibration to above-ground biomass and N concentration resulted in certain patterns for growth-related parameters. The radiation-use efficiency decreased with age and after cutting (first year > second year and first cut > second cut; Figure 1), and increased with nitrogen fertilization, reflecting a positive effect of increased leaf N concentration on the utilization of intercepted radiation (Figure 2). The fraction of biomass allocated to roots decreased with growth period, and the regrowth ability responded similarly to that of radiation-use efficiency. The initial biomass varied with site, indicating a large variation in over-wintering conditions between sites. N uptake under high N availability was limited by biomass in the first growth period, and by biomass and day length in the second growth period. N-uptake efficiency of soil mineral N under limiting N varied with growth period similarly to that of radiation-use efficiency, but was also strongly site specific (Figure 3).

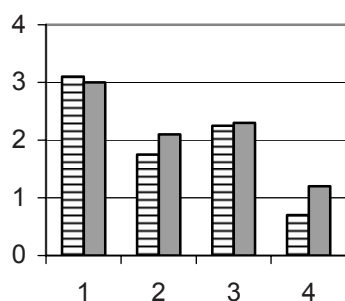


Figure 1. Radiation use-efficiency (g d.w. MJ<sup>-1</sup>; mean over sites and N levels) in relation to first and second growth period in the first year (1, 2) and in the second year (3, 4). Left bars = grass; right bars = mixed clover/grass.

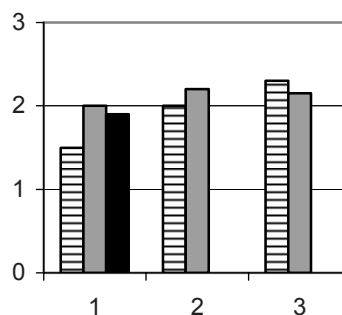


Figure 2. Radiation use-efficiency (g d.w. MJ<sup>-1</sup>; mean over sites and growth periods) for the three nitrogen levels 1, 2 and 3. Left bars = grass; right bars = mixed clover/grass; third bar for 1 is pure clover.

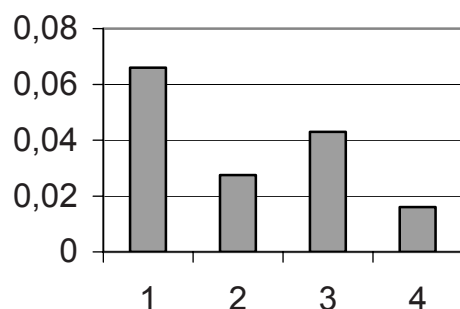


Figure 3. Fraction of soil mineral N in the root zone taken up per day (d<sup>-1</sup>) for non fertilized stands (mean over sites) for the different growth periods (1-4).

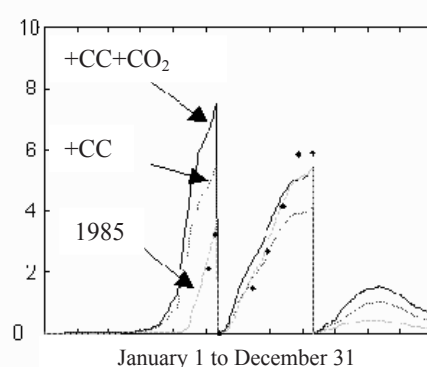


Figure 4. Simulated above-ground biomass (tonnes d.w. ha<sup>-1</sup>) for a fertilized (level 3) grass ley on clay soil in Uppsala for 1985, 1985 + climate change (CC), and CC + increased atmospheric CO<sub>2</sub> concentration, respectively. Points are observed values in 1985.



The climate change assessments showed that the growing period will be extended, and more during the spring than in the autumn. Growth in the spring will start in February in Götaland and Svealand, just over a week later in southern Norrland and in the middle of April in northern Norrland. On fertilized grassland soil water content will decrease from June to October, but growth will not necessarily become lower than the current level, provided unchanged plant N status. Production will increase in the first growth period, which will cause an earlier harvest and more harvests during the season. Introduction of the CO<sub>2</sub> effect was as important as the climate effect, since it made plant more efficient in using both solar radiation and water (Figure 4). The plant-N demand increased considerably more than the availability by increased mineralization, and there was an increased risk of reductions in protein concentrations.

## Conclusions

The model calibration to the field experiments suggested that radiation-use and N-uptake efficiencies of leys decreased after cutting and with age, and that the N-uptake efficiency was strongly site specific. Using the calibrated model for climate change assessments suggested that an additional quantity of irrigation water, corresponding to about the decrease in summer rainfall, and a substantial additional amount of N fertilizers, may need to be applied in order to fully utilize the increased growth potential. However, the variation between sites was high, although only a few site conditions were examined.

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# Influence of root rot on the sustainability of grass/legume leys in Sweden

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## Abstract

Red clover (*Trifolium pratense* L.) is the most important legume crop in Swedish forage production. The *Fusarium* root rot complex is widely prevalent and a major cause of poor persistence of red clover. In a field experiment, the sustainability, yield and quality of two red clover varieties were compared with white clover, lucerne and birdsfoot trefoil in mixed swards with timothy and meadow fescue during three harvest years (2005–2007). The prevalence of root rot was determined twice a year, in spring and late autumn. The results show that root rot was already prevalent in autumn of the establishment year, and after two years all red clover plants investigated were infested. Although the disease severity index was high, the legume content was largest in the red clover treatments during the first two years, but decreased significantly in the third year. Disease level of the other legumes was considerably lower throughout the years.

Keywords: *Fusarium* root rot, red clover, white clover, lucerne, birdsfoot trefoil, organic production

## Introduction

Red clover (*Trifolium pratense* L.) is the most important legume in organic forage production in Sweden. Compared with pure grass leys, red clover contributes to greater dry matter (DM) intake and thereby a larger milk yield (Bertilsson *et al.*, 2001). However, red clover is infested by several fungal soil-borne pathogens affecting its growth, persistence and overwintering capacity, the most important being the root rot complex caused by *Fusarium* spp. and *Cylindrocarpon destructans*. A survey undertaken in 2003–2004 showed damage caused by the root rot complex to be widely distributed on organic farms in south and central Sweden (Wallenhammar *et al.*, 2005). However, other legumes such as white clover (*Trifolium repens* L.) have shown greater persistence than red clover (Svanäng and Frankow-Lindberg, 1994). Lucerne (*Medicago sativa* L.) and birdsfoot trefoil (*Lotus corniculatus* L.) are tolerant of drought, while birdsfoot trefoil is similar to white clover in taste and liveweight gain in growing cattle (Nilsson-Linde *et al.*, 2004). Chicory (*Cichorium intybus* L.) has strong regrowth and an ability to extract nitrogen from deeper soil layers.

The aim of this study was to investigate how to ensure growth and improve quality of ley fodder, in a long-term perspective, by determining the content of legumes in a three-year ley and investigating the occurrence of root rot.

## Materials and methods

A three-year field experiment was established in 2004 on a sandy loamy till (organic matter content 54 mg g<sup>-1</sup> soil) at Rådde Field Research Station in south-western Sweden (57°36'N, 13°14'E, 975 mm mean annual rainfall and 6.1 °C mean annual temperature). An additional experiment with the same treatments was set up in central Sweden (results not shown). A ran-

domized complete block design with four replicates was applied. Red clover (RC) 8 kg ha<sup>-1</sup>, white clover (WC) 4 kg ha<sup>-1</sup>, birdsfoot trefoil (BT) 11 kg ha<sup>-1</sup> or lucerne (LU) 14 kg ha<sup>-1</sup>, were established with seed mixtures containing 10 kg ha<sup>-1</sup> timothy (*Phleum pratense* L.) and 7 kg ha<sup>-1</sup> meadow fescue (*Festuca pratensis* L.). In an additional treatment (D), 1 kg ha<sup>-1</sup> chicory (CH) was added with the RC. The swards were sown with spring barley as a nurse crop harvested in early September. The plots were cut twice or three times a year (Table 1). All treatments were fertilized with 25 Mg ha<sup>-1</sup> of liquid manure in early spring of the second and third year to maintain potassium levels. At each harvest, six samples of about 1 dm<sup>2</sup> from each plot were taken randomly to determine the botanical dry matter (DM) composition. To determine the degree of infestation of root rot, 10 legume plants were dug out randomly from each plot, rinsed and dissected. The internal root symptoms were determined as degree of discolouration in the vascular tissues following Rufelt (1986), and the disease severity index (DSI) was determined. Statistical analysis was performed using SAS Mixed Model.

## Results and discussion

Results of DM yields and content of sown legumes are presented in Table 1. The proportion of red clover DM in RC treatments was significantly higher than the content of sown legume in the other treatments during the first two years. Over the years, the total annual DM yield was largest in RC in the second year, and in WC, BT and LU in the third year (Table 1). There were no differences between RC varieties during the first two years, but in the third year, yield of RC cv. Vivi was larger than that of cv. Fanny. The DM yield in the first year was larger in BT than in WC and LU. Although the proportion of lucerne was already low in the first year, the total yield in the third year was large (9,240 kg ha<sup>-1</sup>) due to the presence of 520 and 310 g kg<sup>-1</sup> volunteer white clover in the second cut in the second and third years, respectively.

Table 1. Means of dry matter (DM) yield (kg ha<sup>-1</sup> and values relative to cv. Fanny) and content of sown legume (Leg., g kg<sup>-1</sup>) in the first, second and third years of mixed timothy-meadow fescue-legume swards with different nos. of cuts (red clover-RC, white clover-WC, birdsfoot trefoil-BT and lucerne-LU and chicory-CH).

Treatment	No. of cuts	Year 1			Year 2			Year 3		
		DM yield		Leg.	DM yield		Leg.	DM yield		Leg.
		kg ha <sup>-1</sup>	Rel	g kg <sup>-1</sup>	kg ha <sup>-1</sup>	Rel	g kg <sup>-1</sup>	kg ha <sup>-1</sup>	Rel	g kg <sup>-1</sup>
A. RC cv. Fanny	2	7740	100	400	9880	100	580	8430	100	230
B. RC cv. Vivi	2	7510	97	330	9900	100	570	9240	110	270
C. RC cv. Fanny	3	7280	94	360	9210	93	510	8670	103	200
D. RC cv. Fanny + CH	3	7110	92	320	9380	95	500	8390	100	210
E. WC	3	5560	72	100	8380	85	260	9120	108	220
F. BT	2	6290	81	100	7830	79	190	8790	104	130
G. LU	3	5550	72	70	8040	81	50	9240	110	50
N		28		28	28		28	28		25
Prob		0.0001		0.0001	0.0001		0.0001	0.0071		0.0282
LSD 0.05		420		70	680		110	520		120

As shown in Figure 1, root rot was prevalent already in autumn the sowing year. The infestation level in red clover was significantly higher (DSI ranged from 35 to 65) than in the other legumes investigated from the second year ( $P < 0.0001$ ), and after two years all of the red clover plants investigated were infested. The DSI of the other legumes was considerably lower, ranging from 6 to 33. However DSI of lucerne was significantly higher than of birdsfoot trefoil and white clover. Despite the high DSI, the content of legumes was largest in the RC

mixtures during the first two years, but decreased significantly in the third year. It was found that secondary roots had developed, as the taproot in red clover was severely damaged. The clover content in RC decreased and that in WC increased so that the levels were similar in the third year. Svanäng & Frankow-Lindberg (1994) found also yields in WC and RC to be similar during the second year and with a larger yield of WC in the third year. The content of sown legumes was larger in the regrowth than in the first cut in all three years, especially in birdsfoot trefoil, as reported by Nilsdotter-Linde *et al.* (2004).

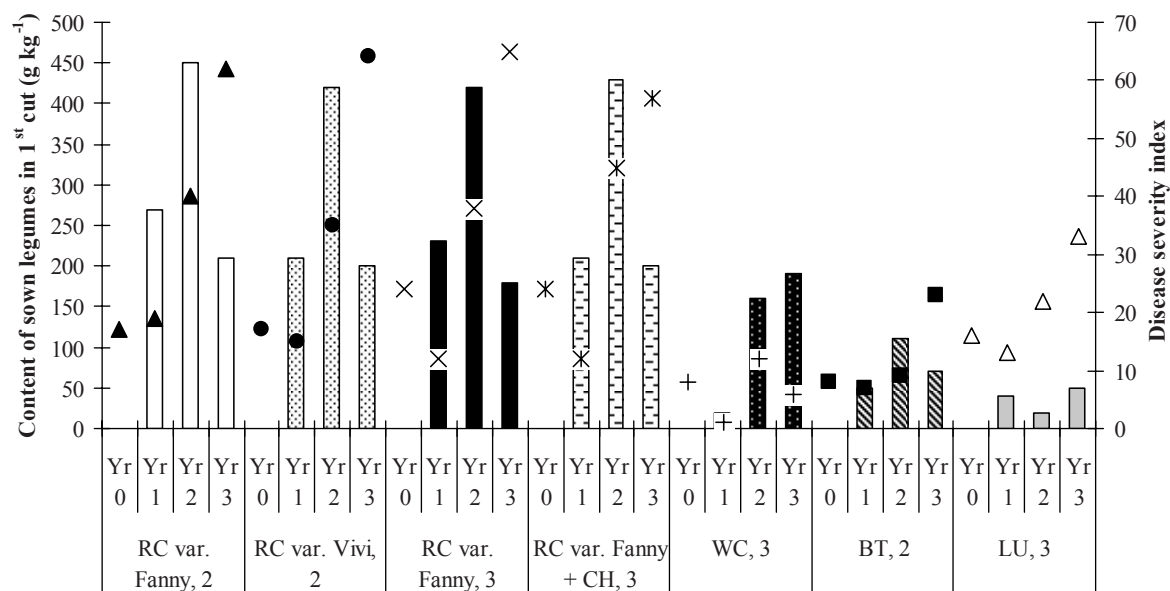


Figure 1. Means of dry matter (DM) content of sown legumes ( $\text{g kg}^{-1}$ ) in mixed timothy-meadow fescue-legume (and chicory) swards according to botanical analysis of the first cut over three years (bars) and disease severity index (DSI) (dots) of roots sampled in October in the sowing year and in April in each year of harvest. See text for explanation of abbreviations.

## Conclusions

Red clover was persistent throughout the second year, although the roots were severely affected by *Fusarium* root rot. During the third year the red clover content significantly declined and the largest DM yield was produced by RC cv. Vivi. To improve the persistence of red clover swards during the third year, access to varieties with some degree of resistance to the root rot complex is required. Furthermore, red clover should be tested for three years in the official variety testing programme.

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## Persistency of *Lolium perenne* in sward under 10-year pasture utilization at varied wetness of peat-muck soil

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### Abstract

An objective was to evaluate persistency of *Lolium perenne* in a permanent pasture sward under conditions of varied wetness on a peat-muck soil (Mt II) during the vegetation season. Mixtures of *L. perenne*, *Phleum pratense*, *Dactylis glomerata* and *Trifolium repens* were sown onto 40 m<sup>2</sup> plots in a randomized block design. Observations of *L. perenne* in the post-boggy habitat over a period of several years confirmed that the most reliable indicator depicting the presence of this species was its share of the herbage in the third regrowth of sward. On the studied pasture, during the third regrowth of sward (June/July), great fluctuations in ground water level were noted. Their effect was manifested by a smaller proportion of *L. perenne* in the sward. Both flooding and a low ground water table appeared to be the reason for a decreased ryegrass share in the pasture sward. The results indicate a relationship between an increasing depth of ground water level in July and *L. perenne* content in the sward, characterized by a correlation coefficient ( $r = 0.65$ ), computed for the compared values throughout the research period.

Keywords: *Lolium perenne*, persistency, pasture, ground water level, peat-muck soil

### Introduction

Persistency of grass species in a plant association on peat-muck soil is a determinant of sward quality and performance, and it also counteracts grassland degradation. However, substantial changes of thermal conditions, and in soil wetness variation in a post-boggy habitat, induce the loss of valuable species of grasses and legumes from the sward.

Perennial ryegrass (*Lolium perenne* L.) is considered a valuable component of pasture sward on mineral soils. On peat-muck soil this species produces rank growth, yet it is susceptible to damage under thermal stresses in winter (Kulik *et al.*, 2004; Warda, 2005). At the same time, it manifests a high capacity for regeneration, especially when provided with sufficient wetness and nitrogen availability (Oomes, 1997). Years of observing *L. perenne* behaviour in the sward under post-boggy habitat confirmed that the most reliable indicator of the presence of this species is its share of the sward in the third regrowth. After the evaluation date of the third regrowth, there is usually sward regeneration after winter damage. At that time it is also possible to determine a relationship between varying wetness of peat-muck soil and persistency of *L. perenne* in the sward. Great fluctuations in ground water level over the vegetation period may hamper ryegrass development and its persistency.

One of the objectives of these studies was to evaluate *L. perenne* maintenance in the permanent pasture sward under the conditions of varied wetness of a peat-muck soil.

### Materials and methods

The investigations initiated in 1996 by Krzywiec (2000) are being continued. The experiment was set up on peat-muck soil (Mt II) with a randomized blocks design and four replications. The mixture of grasses (65%) and white clover (*Trifolium repens*) (35%) was sown onto plots of 40 m<sup>2</sup> area. The grass seed proportions in the total mixture were *L. perenne* (35%), *Phleum*



*pratense* (20%) and *Dactylis glomerata* (10%). A pure grass mixture was also sown, composed of the above-mentioned grass species, and the share contributed by white clover in the grass-clover mixture was distributed proportionally between the grasses constituting the mixture, so that *L. perenne* accounted for 54% of the mixture. In the years of full pasture utilization, the grass sward was fertilized with nitrogen at amounts of 40, 80 and 120 kg ha<sup>-1</sup>, while the grass-white clover sward with 40 kg N ha<sup>-1</sup> year<sup>-1</sup>. Equal amounts of phosphorus and potassium was applied all over the grassland area, at 35 kg P and 100 kg K ha<sup>-1</sup> year<sup>-1</sup>. Four grazings by Limousin cattle were conducted over the grazing season. Throughout the year (four times a month), measurements of a ground water level were taken in the well situated at the experimental area. Weather conditions were recorded by the automatic meteorological station (ASM-971). Before the animals were admitted to the pasture quarter, the sward was sampled to determine dry matter yield and afterwards the sward species composition was determined using the botanical-weight method. Relationship between the depth of ground water level and *L. perenne* content in the sward throughout the research period was determined by calculating the obtained data according to the arstat.core programme (prepared in Information Technology Centre of University of Life Sciences in Lublin).

## Results and discussion

The highest fluctuations of *L. perenne* content were recorded in the grass-clover sward (Table 1), yet the average value characterizing ryegrass presence in this sward type throughout the experimental period was substantially greater than its species share in the mixture at the time of sowing.

Table 1. Average share of *Lolium perenne* (% DM) in the third sward regrowth depending on the sward type and nitrogen (N) fertilization (kg ha<sup>-1</sup>), in the years 1996-2006.

Treatment	Ranges of content	Mean
Grass-clover sward (G-C) + N <sub>40</sub>	24.5-89.1	51.2
Grass sward (G) + N <sub>40</sub>	30.2-78.8	55.1
Grass sward (G) + N <sub>80</sub>	40.8-86.4	67.4
Grass sward (G) + N <sub>120</sub>	47.0-81.9	67.7

The proportion of *L. perenne* in the grass sward showed minor fluctuations with increasing nitrogen dose. Analysing the mean values, it was found that the quantity of this species in the 40 kg N ha<sup>-1</sup> treatment was close to its share in the mixture. In the pasture area fertilized with 80 and 120 kg N ha<sup>-1</sup>, ryegrass behaved similarly and appeared to be the dominant species. However, such a high content of *L. perenne*, a species susceptible to thermal stress in the winter period, is not beneficial to the persistency of pasture association in the post-boggy habitat (Warda, 2005).

On the studied pasture, the third regrowth of sward was obtained at the end of June/early July, and at this period – especially in the years 1997 (18.6 cm), 1998 (76.7 cm), 2003 (79.2 cm) and 2006 (79.7 cm) – great fluctuations in ground water level were noted. Their effect was manifested by a smaller proportion of *L. perenne* in the sward of this regrowth (Figure 1). Flooding of the area and low ground water table appeared to be the reasons for a decrease of ryegrass share in the pasture sward. The research results obtained here indicate a dependence between an increasing depth of a ground water level in July and *L. perenne* content in the sward, characterized by a correlation coefficient ( $r = 0.65$ ), computed for the compared values throughout the research period.

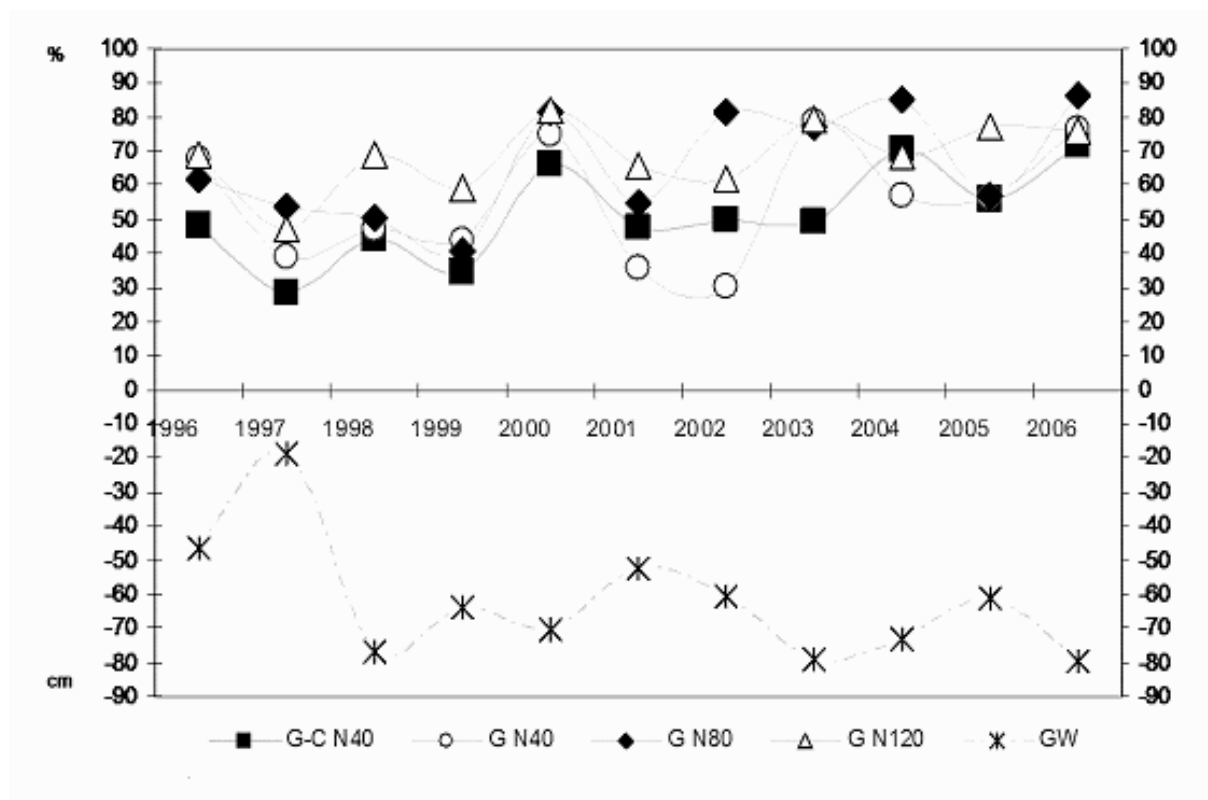


Figure 1. Relationship between the content of *Lolium perenne* and the ground water (GW) level during the third regrowth of pasture sward in the years 1996-2006.

## Conclusions

The most reliable indicator depicting the presence of *L. perenne* in the sward is its share in the third regrowth. The effect of habitat conditions is very important for the persistency of *L. perenne* in permanent pasture on peat-muck soil. An increasing depth of ground water level in July affected the quantity of *L. perenne* in the sward under a 10-year pasture utilization period. Fluctuations of *L. perenne* content depended on the sward type, and the greatest ranges in it share of the sward were obtained in the grass-clover sward.

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# Effect of N fertilization on the development of couch-grass (*Elytrigia repens*) in a grass sward

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## Abstract

The effect of N fertilization on the development of couch-grass (*Elytrigia repens*) was investigated in a 4-year small-plot trial. The experiment was situated at 390-402 m above sea level in the Hruby Jeseník Mountains. There were four methods of utilization: intensive (4 cuts per year), moderately intensive (3 cuts per year), low intensive (2 cuts per year), and extensive (2 cuts per year), and four levels of fertilization: A – without fertilizer; B – P<sub>30</sub>K<sub>60</sub>; C – N<sub>90</sub>P<sub>30</sub>K<sub>60</sub> and D – N<sub>180</sub>P<sub>30</sub>K<sub>60</sub>. The dominant grass species in the first year were *Poa pratensis*, *Dactylis glomerata* and *Lolium perenne*. In the following years there was a significant increase of *Elytrigia repens* in the treatments with N fertilization. By the statistical LSD test of the projective dominance, significant ( $P < 0.05$ ) differences were found among individual fertilization treatments. There was a direct correlation between development of *Elytrigia repens* and higher levels of N fertilization.

## Introduction

Meadows are secondary successional formations which have developed under human impact through the systematic removal of shoot biomass (grazing and mowing). Mowing is the basic management attribute for the existence of meadows. Meadows have evolved to be adapted to seasonal changes in management (Blažková 1989), and fertilization is also part of the management of meadows. The influence of fertilization on plant species diversity is immediate and very significant. The aim was to investigate the effect of N fertilization on the development of couch-grass (*Elytrigia repens*) in a grass sward. A similar theme was considered by Tardif and Leroux (2002) and Malhi *et al.* (2003).

## Materials and methods

A small-plot trial was established in 2003 on permanent grassland sites with a range of cutting intensities and various levels of the mineral fertilization in the locality of Rapotín, Hruby Jeseník Mountains (Czech Republic). The dominant botanical species at the beginning of trial were *Poa pratensis*, *Dactylis glomerata*, *Lolium perenne*, *Trifolium repens* and *Taraxacum sect. Ruderalia*. The experiment was located on an east-facing slope (of 5.1-6.2°) at an elevation of 390-402 m above sea level. The soil was considered to be a cambisol (horizons Ao-Bv-B/C-C) with characteristics of sandy-loam.

Table 1. Mean seasonal and annual precipitation (mm) and air temperatures (°C).

Year	Sums of precipitation (mm)				Mean air temperatures (°C)			
	2003	2004	2005	2006	2003	2004	2005	2006
Per growing season	248.1	272.4	315.8	373.0	15.0	13.7	14.1	14.9
Per year	546.3	617.8	603.5	667.8	6.0	7.3	7.3	7.8

The experiment consisted of 16 treatments (four levels of mineral fertilization and four levels of cutting frequency) with four replicates. The fertilization treatments were: A – without

fertilization, B – P<sub>30</sub>K<sub>60</sub>, C – N<sub>90</sub>P<sub>30</sub>K<sub>60</sub>, and D – N<sub>180</sub>P<sub>30</sub>K<sub>60</sub>. The treatments representing levels of cutting utilization were: intensive (4 cuts per year, first cut on 15 May followed by cuts at 45d-intervals); moderately intensive (3 cuts per year, first cut on 30 May followed by cuts at 60d-intervals); low intensive (2 cuts per year, first cut on 15 June and a further cut after 90 days); and extensive (2 cuts per year, first cut on 30 June with a further cut after 90 days).

Before the first cut, botanical analysis was assessed by the method of reduced projective dominance (Regal and Veselá, 1975) for the purpose of determining changes in botanical composition of grassland. The evaluation of the influence of the N fertilization by couch-grass was carried out by means of the statistical method, LSD test.

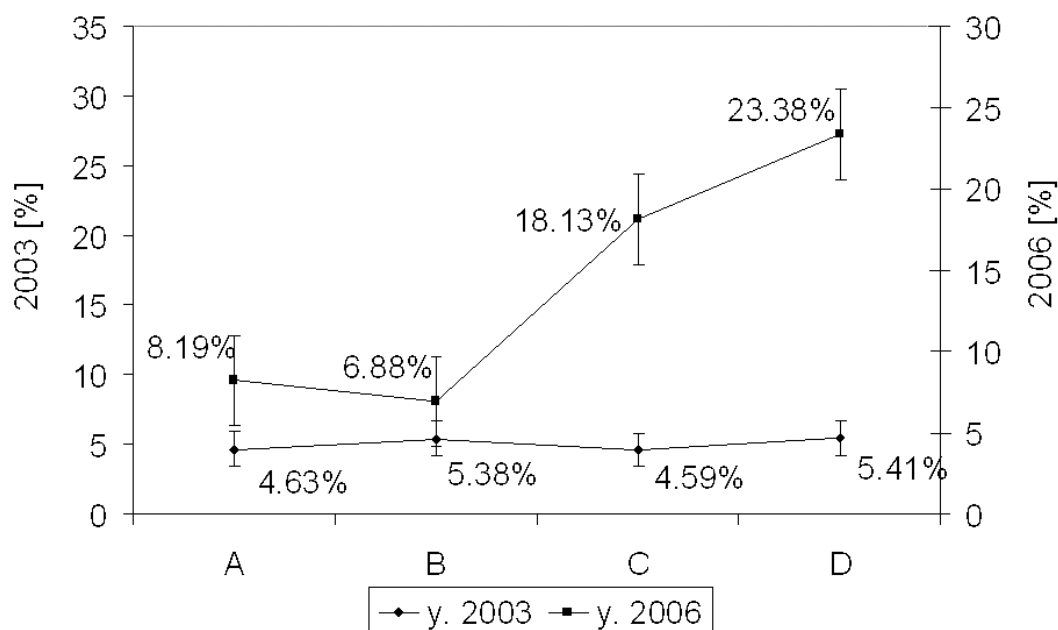
## Results and discussion

The dominance of agrobotanical groups in relation to fertilization is shown in Table 2.

Table 2. Relative proportions (as %) of agrobotanical groups for each fertilization treatment in years 2003 and 2006.

Year	2003				2006			
Treatment	A	B	C	D	A	B	C	D
Herbs(%)	32.1	29.4	26.1	25.2	52.0	45.9	39.3	27.1
Grasses(%)	55.3	54.4	60.7	66.4	41.1	42.3	57.7	71.3
Legumes(%)	12.6	16.2	13.2	8.4	6.9	11.8	3.1	1.5

There was a significant increase of native species and decrease of legumes under the treatment without fertilization (A). Nitrogen fertilization increased the dominance of grasses and decreased legumes in the plant cover (D). This corresponds with the results published by other authors including Klimeš (1999) and Hrabě *et al.* (2004).



Vertical bars denote 0.95 confidence intervals

Figure 1. The % cover of couch-grass (*Elytrigia repens*) in 2003 and 2006 in relation to fertilization treatment.

Among the individual variants of fertilization there were statistically significant differences between the following treatments: A – C, A – D, B – C, B – D and C – D.

At the start of the experiment there was a relatively high cover of rhizomatous grasses such as *Poa pratensis* and *Elytrigia repens*. After regular doses of fertilization there was a significant increase of *Elytrigia repens* (Figure 1) in the treatments with nitrogen fertilization (C and D). Oerlemans *et al.* (2005) came to the same conclusion from his trial on the long-term effect of N-fertilization. The occurrence of this species in meadow stands for hay production is tolerated in amounts of 12-15% (Hrabě and Buchgraber, 2004). The results of statistical tests are summarized in Figure 1. The LSD test showed significant differences ( $P < 0.05$ ) between the beginning and the end of the experiment in coverage of the weed grass *Elytrigia repens*. The confidence interval at significance level  $P < 0.05$  was 1.24 in 2003 and 2.77 in 2006.

## Conclusions

The four years of fertilization brought changes in the phytocoenological composition of the vegetation. It showed the influence of nitrogen fertilization on increasing the cover of rhizomatous grasses in the vegetation and the significant increase of *Elytrigia repens* under the treatments with nitrogen fertilization. The higher doses of N fertilization increased the proportion of couch-grass in the sward and to avoid excessive increases in this species it is better to apply N only in doses up to 90 kg N ha<sup>-1</sup> y<sup>-1</sup>. This may be considered suitable for those areas that have similar natural conditions to this trial site.

## Acknowledgements

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# The productivity of early type red clover (*Trifolium pratense*) varieties and breeding lines

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## Abstract

Red clover varieties and breeding lines were tested at the Lithuanian Institute of Agriculture in Dotnuva in 2005 and 2006. The Lithuanian varieties 'Liepsna', 'Vyčiai', 'Vyliai', and newly developed breeding lines and varieties received from foreign scientific institutions (8 varieties and 10 breeding lines altogether) were investigated. Plant height, dry matter yield, and seed production were recorded. The diploid variety 'Vyčiai' was chosen as a standard. All varieties and breeding lines were early maturing. The tetraploid variety 'Sadūnai' was noted for its plant height (61.6 cm), but its seed production was lower ( $6.7 \text{ kg ha}^{-1}$ ) than that of the standard diploid variety. The variety 'Astra' performed worst of all the varieties tested, its height was 51.2 cm and dry matter yield was  $7.38 \text{ Mg ha}^{-1}$ . The highest yielding variety was 'Radviliai' and its dry matter yield was  $1.68 \text{ Mg ha}^{-1}$  more than that of the standard variety. The diploid breeding line No 33 exhibited the highest seed production, while the variety 'Vyliai' had the lowest seed production ( $260 \text{ kg ha}^{-1}$  and  $106.7 \text{ kg ha}^{-1}$ , respectively).

Keywords: red clover, yield, seed production

## Introduction

Increasing intensity of forage production has boosted the demand for high-yielding, disease-resistant legumes that exhibit good winter survival, high feeding value and seed set (Barker and Kalton, 1989). Under Lithuania's conditions red clover (*Trifolium pratense* L. ( $2n = 14$ )) is one of the key legumes grown for forage. It is a productive perennial plant which has low seed productivity (Svirskis, 1995; Dabkevičienė, 2002). Satisfactory clover seed production is determined by a sufficient amount of rainfall before flowering and sunny dry weather during flowering and seed ripening. A protracted flowering and ripening period can exert a negative impact.

Two types of red clover (early and late) are grown in Lithuania. When grown for seed, early clover ripens more rapidly than late clover. Early red clover (*Trifolium pratense*, var. *Foliosum* Choc.) grows faster, and starts flowering earlier than late clover, and produces two herbage yields. If the first cut is taken at the right time, seed can be expected to ripen in the second crop (Lazauskas, 1998).

The aim of the investigation was to establish productivity in red clover varieties and newly created breeding lines.

## Materials and methods

Red clover varieties and breeding lines were tested in 2005 and 2006 at the Lithuanian Institute of Agriculture (Dotnuva) on a medium heavy gleyic cambisol (CMg). The Lithuanian varieties ('Liepsna', 'Vyčiai', 'Radviliai'- diploid, 'Vyliai', 'Sadūnai'- tetraploid), the varieties received from foreign scientific institutions ('Brita', 'Astra', 'Sabtoron'- diploid) and newly created breeding lines (33, 2288, 2271, 2297, 2270, 2299 and 2182 – diploid; 2300, 2301, 2286 – tetraploid) were investigated. Plant height, dry matter yield and seed production were assessed. The diploid variety 'Vyčiai' was chosen as a standard. All varieties and breeding lines were early maturing. The genotypes were planted in  $5.5 \text{ m}^2$  test plots on

1 June 2004 without cover crop, using a randomized complete block design with five replications. Phosphorus and potassium fertilizers (P<sub>60</sub> K<sub>90</sub>) were applied in the autumn of each year of use.

In 2004 the spring period was warm and dry. In June the weather was cool and there was less rainfall than the long-term average. Warm and rainy weather dominated in the second-half of summer. In 2005 the weather conditions were changeable in terms of air temperature: warmer periods alternated with cooler ones. The greatest amount of rainfall fell at the beginning of June, end of July and beginning of August. The autumn was warm and long. At the beginning of winter the conditions were favourable for red clover. In 2006 the winter period was cold, long, with a permanent thin snow layer. The spring was cold, dry and long. The summer was warm and dry. July was the warmest month, while the driest month was June, and the rainiest month was August. During the greater part of summer the reserves of productive moisture in the soil for plants were critical. Beginning of September was warm and rainy; later the weather conditions turned to dry and sunny (Table 1).

Table 1. Precipitation and temperature data (April-October) for in Central Lithuanian region (Dotnuva) for the study period (2004-2006) with long-term (1924-2006) average.

Month	Temperature (°C)				Precipitation (mm)			
	Average			Long-term 1924-2006	Average			Long-term 1924-2006
	2004	2005	2006		2004	2005	2006	
April	7.6	7.6	6.7	5.7	11.1	23.9	19.2	37.4
May	11.2	12.4	12.6	12.2	27.8	46.1	45.0	52.0
June	14.2	15.3	16.8	15.6	44.2	50.3	6.8	61.2
July	16.9	19.3	21.3	17.6	81.6	46.3	40.4	73.0
August	18.1	16.8	18.1	16.6	94.5	75.5	105.0	73.8
September	12.9	14.4	14.6	12.0	53.2	26.6	76.0	51.8
October	8.2	7.9	9.7	6.8	68.8	20.5	49.2	49.3

The experimental data were developed by P. Tarakanovas computer software 'YIELD' and 'STAT ENG' (this program includes correlation analysis) in the Visual Basic Application as a macro program to run in EXCEL was used.

## Results and discussion

Red clover dry matter, seed production and plant height depends on the weather conditions, and they were not favourable during the experimental years. It was warm enough, but there was a lack of humidity during plant growth and flowering. The highest yielding variety of all varieties investigated was 'Radviliai': its dry matter yield was higher by 1.68 Mg ha<sup>-1</sup> than that of the standard variety 'Vyčiai'. This variety also exhibited satisfactory seed production (223.3 kg ha<sup>-1</sup>) and the plants were high (60.9 cm).

The variety 'Sadūnai' was noted for mean plant height (61.6 cm) and it produced the highest total dry matter yield (9.46 Mg ha<sup>-1</sup>) and best seed production (203.3 kg ha<sup>-1</sup>) among all tetraploid varieties and breeding lines investigated. The breeding line No 2286 and variety 'Vyliai' produced the highest dry matter yield among the tetraploid varieties and breeding lines (9.97 Mg ha<sup>-1</sup> and 9.85 Mg ha<sup>-1</sup>), but their seed production was low (110.0 kg ha<sup>-1</sup> and 106.7 kg ha<sup>-1</sup>). The variety 'Astra' performed worst of all the varieties tested; its height was 51.2 cm, and dry matter yield was 7.38 Mg ha<sup>-1</sup>. However, this variety produced the greatest seed yield at 226.3 kg ha<sup>-1</sup>. The diploid breeding line No 33 exhibited the highest seed production (260 kg ha<sup>-1</sup>). Plants of the tetraploid varieties and breeding lines were higher, and produced a greater dry matter yield than diploid ones. However, diploid varieties

demonstrated better seed production, except for the Lithuanian tetraploid variety 'Radviliai' (Figure 1).

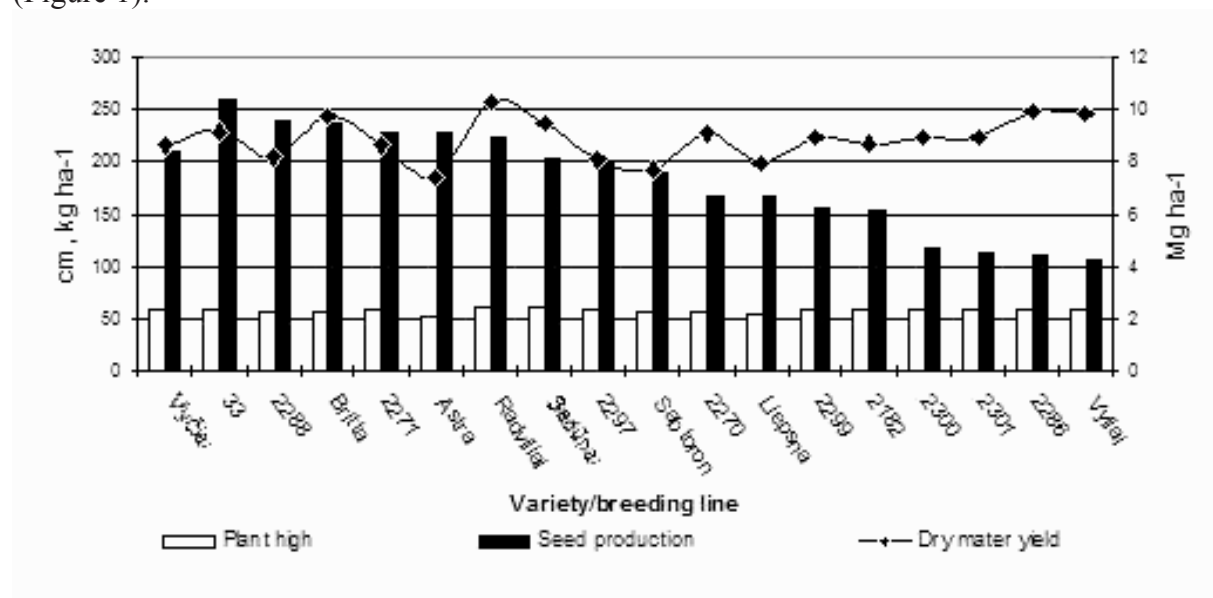


Figure 1. Mean plant height, total dry matter yield and seed production of early type red clover varieties and breeding lines, 2005-2006.

The correlation analysis for the investigated characters showed average positive correlation ( $P < 0.01$ ) between plant height and dry matter yield ( $r = 0.66$ ). The correlation between seed production and plant height and dry matter yield was not established (Table 2).

Table 2. The correlations between red clover mean plant height, total dry matter yield and seed production. Dotnuva 2005-2006. (\*\* significant at  $P < 0.01$ )

Character	Dry matter yield	Seed production
Plant height	0.66**	-0.169 n
Dry matter yield		-0.266 n

## Conclusions

Tetraploid varieties and breeding lines produced better dry matter yield, and their plants were taller than those of diploid varieties, but diploids exhibited better seed production, except for the Lithuanian tetraploid variety 'Sadūnai'. The diploid variety 'Radviliai' and tetraploid variety 'Sadūnai' were distinguished by satisfactory yielding among the varieties tested. The variety 'Astra' had high seed production. Positive correlation was established between plant height and dry matter yield. The results indicate that continuation of red clover breeding using both diploid and tetraploid varieties could be very promising.

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# Performance of silage maize in forage crop rotation systems

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## Abstract

A field experiment with three years of measurements was carried out at a typical site for forage production in northern Germany in order to study the effects of different crop rotation systems on yield and quality of forage maize (*Zea mays* L.) within the systems. Two three-year forage crop rotation systems (silage maize – winter wheat for whole crop silage – winter barley for whole crop silage; silage maize – two years forage grasses) were compared with silage maize in each year (monoculture with and without grass understorey; overwintering forage grasses with the first cut in spring). Three levels of fertilization (0, 110, 160 kg N ha<sup>-1</sup>) were applied, including two levels of slurry (0, 80 kg organic N ha<sup>-1</sup> y<sup>-1</sup>). Nitrogen effects were shown as linear or quadratic regressions. The results showed, on average over three years, no differences in yield and quality parameters between mineral and slurry nitrogen and the highest dry matter yield, net energy (NE<sub>L</sub>) yield and NE<sub>L</sub>-concentration for silage maize in rotation with cereals for whole crop silage, especially with low nitrogen input. Maize after the first cut of forage grasses had only 85% of the productivity of maize in monoculture.

Keywords: forage maize, maize yield, maize quality, forage crop rotation, nitrogen fertilization, whole crop silage

## Introduction

Forage production on the sandy soils in northern Germany is based mainly on herbage from grassland and silage maize in monoculture. Maize production in monoculture is associated with several problems, such as soil erosion, loss of organic matter and nitrogen losses, especially during the vegetation-free period of the year. Adapted nitrogen fertilization to maize (Wulfes *et al.*, 2000) or maize production in crop rotation systems (Wachendorf *et al.*, 2004) are suitable techniques for increasing nitrogen efficiency and can reduce nitrogen leaching out of the system (Wachendorf *et al.*, 2006). In this investigation the effects of different crop rotation systems on yield and quality of forage maize for a typical sandy soil location in northern Germany were examined.

## Materials and methods

Two three-year forage crop rotation systems (i) silage maize – winter wheat for whole crop silage – winter barley for whole crop silage, followed by summer rape (*Brassica napus* L.) as a catch crop over winter, and (ii) silage maize, followed by two years of forage grasses, were compared with three maize production systems, where silage maize is grown in each year: monoculture (iii) with and (iv) without grass understorey and (v) alternation of maize and overwintering forage grasses with a first cut in early spring. The field experiment was carried out in a split-plot design with 3 replicates from 2002 to 2004 with a preceding year for establishing all crops. In the three-year rotation systems each crop grows in each year. The plot size was 3 x 10 m.

The mean N-fertilization levels of all crops in rotation were 0 kg N ha<sup>-1</sup> y<sup>-1</sup> (extensive level), 160 kg N ha<sup>-1</sup> y<sup>-1</sup> (reduced level), and 220 kg N ha<sup>-1</sup> y<sup>-1</sup> (optimal level) with special nitrogen amount for each crop, including two levels of slurry (0, 80 kg organic N ha<sup>-1</sup> y<sup>-1</sup>). N-appli-



cations for maize were 0, 110 and 160 kg N ha<sup>-1</sup> y<sup>-1</sup>, including two levels of slurry (0, 80 kg organic N ha<sup>-1</sup> y<sup>-1</sup>). Slurry was applied before sowing of maize.

In addition to the yield parameters of dry matter (DM) yield, net energy (NE<sub>L</sub>) yield, nitrogen (N) yield and starch yield, quality parameters DM-content, NE<sub>L</sub>-content, crude protein (CP) content and starch content were measured by NIRS. Because the interaction of crop rotation system (CRS) x slurry nitrogen was not significant, the effects of all nitrogen levels (mineral and organic) were shown as linear or quadratic regressions.

## Results and discussion

The crop rotation system had significant influence on all investigated parameters (Table 1). In the mean of all nitrogen levels, including the slurry application, the maize in rotation with winter cereals for whole crop silage exhibited the highest amount of dry matter, net energy, starch and nitrogen, followed by the maize in monoculture without grass understorey and the maize after two years of forage grasses. Maize in each year with overwintering forage grasses and a first cut in early spring resulted in a 15% lower yield than the maize in monoculture without any crops over winter. Differences of 3.7 Mg DM ha<sup>-1</sup>, 23.2 GJ NE<sub>L</sub> ha<sup>-1</sup>, 2 Mg starch ha<sup>-1</sup> and 62 kg N ha<sup>-1</sup> in maize yield indicated that there was a large influence of the crop rotation system on productivity of maize for silage (Wachendorf *et al.*, 2004).

The low DM- and starch-content of the maize after a first cut indicated that the vegetation period for the maize in this system is too short for adequate ripeness, compared with the other maize crops. Otherwise, it must be considered that the first grass cut reached 4 Mg ha<sup>-1</sup> DM additionally to the maize crop. The quality parameters of the maize in all other rotation systems are significantly different but, with exceptions of the maize after first cut of grass and the maize with grass understorey, the differences are of a low level. Maize after 2 years of forage grasses had the highest crude protein content and nitrogen uptake, whereas the starch content was reduced. Grass understorey in a maize crop reduced the CP-content of maize extremely, especially with low nitrogen input, which indicates a great competition between these two crops (Figure 1).

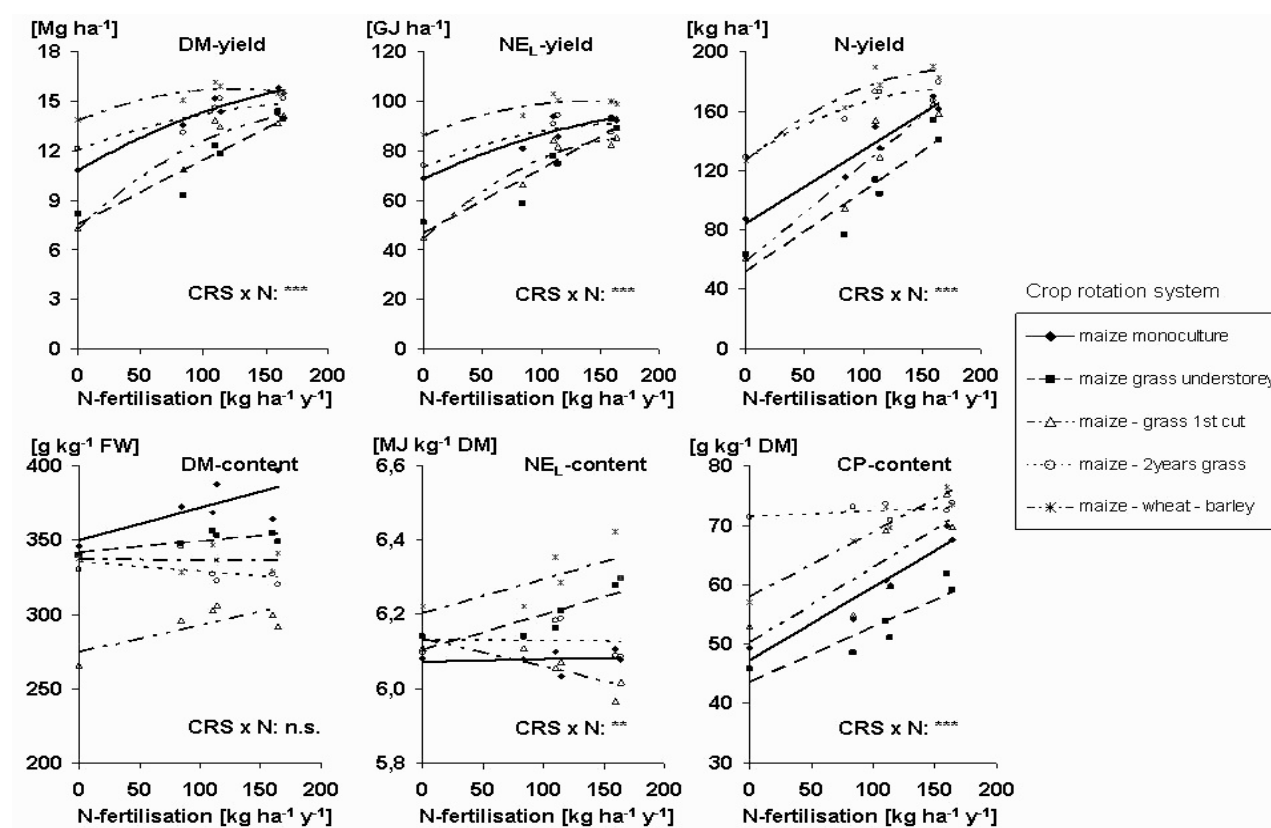
Table 1. Influence of crop rotation system on yield and quality parameters of silage maize (mean of three nitrogen levels, two slurry application rates, three years and three replicates).

Crop rotation system	DM Mg ha <sup>-1</sup>	NE <sub>L</sub> GJ ha <sup>-1</sup>	Starch Mg ha <sup>-1</sup>	N kg ha <sup>-1</sup>	DM g kg <sup>-1</sup>	NE <sub>L</sub> MJ kg <sup>-1</sup> DM	Starch g kg <sup>-1</sup> DM	CP g kg <sup>-1</sup> DM
Maize monoculture	14.21 <sup>b</sup>	85.97 <sup>b</sup>	4.25 <sup>a</sup>	136.6 <sup>b</sup>	372.9 <sup>a</sup>	6.06 <sup>c</sup>	297.4 <sup>a</sup>	60.3 <sup>d</sup>
Maize grass understorey	11.65 <sup>c</sup>	73.99 <sup>c</sup>	3.55 <sup>b</sup>	108.8 <sup>d</sup>	350.0 <sup>b</sup>	6.20 <sup>b</sup>	272.5 <sup>b</sup>	53.4 <sup>e</sup>
Maize – grass 1st cut	12.22 <sup>c</sup>	74.15 <sup>c</sup>	2.47 <sup>c</sup>	127.1 <sup>c</sup>	293.8 <sup>c</sup>	6.11 <sup>bc</sup>	199.1 <sup>d</sup>	63.6 <sup>c</sup>
Maize – 2 years grass	14.60 <sup>ab</sup>	89.92 <sup>b</sup>	3.46 <sup>b</sup>	170.2 <sup>a</sup>	334.6 <sup>b</sup>	6.14 <sup>bc</sup>	235.7 <sup>c</sup>	72.5 <sup>a</sup>
Maize – wheat – barley	15.35 <sup>a</sup>	97.16 <sup>a</sup>	4.49 <sup>a</sup>	171.6 <sup>a</sup>	336.8 <sup>b</sup>	6.30 <sup>a</sup>	286.9 <sup>ab</sup>	69.5 <sup>b</sup>
LSD <sub>0.05</sub>	1.11***	7.16***	0.46***	12.4***	21.9***	0.09***	19.0***	2.7***

DM = dry matter, NE<sub>L</sub> = net energy of lactation, CP = crude protein. LSD = least significant difference with  $P < 0.05$ . Means in the same column with the same superscript letters do not differ at  $P < 0.05$ .

Differences in the yield parameters of maize due to crop rotation systems are mainly based on differences in the low nitrogen level, whereas the quality parameters often differed with high amount of nitrogen (Figure 1). Maize in rotation with cereals or with 2 years of forage grasses, and even in monoculture, showed no significant increasing DM- and NE<sub>L</sub>-yield with

ascending nitrogen level from 110 to 160 kg N ha<sup>-1</sup> y<sup>-1</sup> (Wulfes *et al.*, 2000), whereas maize after the first cut of grass, and maize with grass understorey, reacted with further increasing yield.



DM = dry matter, NE<sub>L</sub> = net energy of lactation, CP = crude protein, N = nitrogen, CRS = crop rotation system, \*\*\* =  $P < 0.001$ , \*\* =  $P < 0.01$ , ns =  $P > 0.05$ .

Figure 1. Yield and quality parameters of silage maize depending on crop rotation system (CRS) and amount of nitrogen fertilization (N = mineral + organic, mean of three years).

## Conclusions

The results reflected the different nitrogen and soil water demands of the different crop rotation systems. Maize in monoculture without a grass understorey, or maize in rotation with cereals or 2 years of forage grasses, has only a small yield reduction with decreasing nitrogen level, whereas maize with a grass understorey or after a first cut in spring suffers under a reduced nitrogen input. For evaluation the whole rotation should be considered.

## Acknowledgements

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# Changes in tillering and leaf chemistry in *Brachypodium phoenicoides* infected by the fungus *Epichloë typhina*

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## Abstract

The fungus *Epichloë typhina* (*Clavicipitaceae*) is the causal agent of choke disease in several grass species. This disease is characterized by the presence of cylindrical fungal stromata which wrap the immature inflorescences and inhibit the normal development of reproductive tillers. A three-year field experiment conducted with infected and uninfected plants of the grass *Brachypodium phoenicoides* showed that in a vegetative growth stage there were no significant differences in biomass production, but the concentration of Ca, Mg and Mn was greater and that of Na was lower in infected than in non-infected plants. Infected plants produced up to twice as many reproductive tillers than healthy plants, but their reproductive tissue biomass was smaller than that of healthy plants, since tiller development was arrested by choking stromata.

## Introduction

*Epichloë* species and their asexual *Neotyphodium* relatives (Fam. *Clavicipitaceae*, *Ascomycota*) form systemic and perennial associations with grasses, ranging from antagonistic to mutualistic (Schardl *et al.*, 2004). *Epichloë typhina* is a pathogenic species which forms a cylindrical fungal stroma around inflorescences when host plants enter into the reproductive growth stage. As a result the panicles cannot emerge and the plant is sterilized. These symptoms are known as choke disease. The disease can cause economic problems in seed crops of some host grasses.

There are numerous studies on growth and physiological effects of mutualistic *Epichloë* or *Neotyphodium* species on their host plants (Schardl *et al.*, 2004). However, little is known about how plants are affected by interactions with pathogenic species, besides being sterilized (Pan and Clay, 2002). The objectives of this study were to investigate the effects of *Epichloë typhina* infection on the growth and nutrient content of the grass *Brachypodium phoenicoides*.

## Materials and methods

Diseased plants of the perennial grass *Brachypodium phoenicoides* were obtained in a meadow in Torres del Carrizal (Zamora, western Spain). At this location 5 to 10% of the plants showed symptoms of choke disease.

A set of infected and uninfected plants of identical genotype was produced to study the effects of choke disease on the vegetative growth of the plants. This set was developed from a single infected plant by dividing it into several ramets. One half of the ramets were treated with a fungicide in order to kill the fungus (Zabalgogazcoa *et al.*, 2006).

In April 2001, six treated and six untreated plants of similar size were transplanted to the field. A space of 60 cm was left between the randomly positioned plants. The soil was an eutric chromic cambisol with neutral pH at the surface, decreasing slightly with depth. To imitate natural conditions, plants were not fertilized during the 40 months of the experiment.

Biomass of the vegetative stage of each plant was determined by cutting plants 3 cm above ground level in November 2001, April 2003, and November 2004. Yield in the reproductive

stage was measured in harvests made in July 2002, 2003, and 2004, when the healthy plants had mature flowering tillers. The number of reproductive tillers was counted for each plant at this time. Plants were not cut or grazed between harvests.

For nutrient element analysis, vegetative-stage samples harvested in April 2003 were used. Leaf material was dried in a forced air oven at 60 °C for 48 hours before weighing and then ground in a Retsch ZM1 mill with a 1 mm mesh sieve. Each sample was analysed for N, P, K, Ca, Mg, Na, Mn, Cu and Zn concentrations using the methods described by Zabalgogezcoa *et al.* (2006). The statistical significance of the differences between means of infected and uninfected plants in terms of biomass, tiller number, or nutrient content were tested with a Student's t-test with  $\alpha = 0.05$ .

## Results and discussion

Most reproductive tillers of infected plants were sterilized by *Epichloë* stromata. In July 2002, the second growing season, 94% of the reproductive tillers had stromata, in 2003 94%, and in 2004 83%.

There were no significant differences ( $P > 0.05$ ) in the N, P, K, Zn or Cu content of leaves collected from infected and uninfected plants in April 2003 (Table 1). However, Ca, Mg, and Mn concentrations were greater ( $P < 0.05$ ) in infected than in healthy plants, and the opposite occurred with Na content ( $P < 0.05$ ). This suggests that *E. typhina* infection may have an effect on the tissue chemistry of its host.

The differences in biomass production between infected and uninfected plants in the vegetative growth stage were not statistically significant ( $P > 0.05$ ) in any of the three years that this parameter was measured. In 2001 the average vegetative plant weight was 34.10 g, in 2003 70.45 g, and in 2004, 24.15 g. Choke disease had a significant effect ( $P < 0.05$ ) on the number of reproductive tillers per plant (Figure 1A). The average number of tillers per plant was greater in infected plants than in uninfected plants during the three years. The difference was statistically significant ( $P < 0.01$ ) in 2003 and 2004 with infected plants producing 1.8 and 2.0 times more reproductive tillers than healthy plants. As a result of the arrested development of flowering tillers, the reproductive biomass was lower in infected than in healthy plants in all three years ( $P < 0.05$ , Figure 1B).

Table 1. Average nutrient content ( $\pm$  s.d.; dry weight measurements from six plants) of leaves of infected and uninfected plants in vegetative stage harvested in 2003. Means of each nutrient were compared by means of a Student's t-test. Statistically significant difference exists between means marked by an asterisks (\* $P < 0.05$ ; \*\* $P < 0.01$ ).

Element	Infected	Healthy
N (g kg <sup>-1</sup> )	25.57 $\pm$ 1.20	25.62 $\pm$ 1.36
P (g kg <sup>-1</sup> )	2.07 $\pm$ 0.44	1.92 $\pm$ 0.07
K (g kg <sup>-1</sup> )	11.45 $\pm$ 2.05	12.90 $\pm$ 1.90
Ca (g kg <sup>-1</sup> )	2.85 $\pm$ 0.10 **	2.42 $\pm$ 0.07 **
Mg (g kg <sup>-1</sup> )	0.73 $\pm$ 0.05**	0.63 $\pm$ 0.05 **
Na (g kg <sup>-1</sup> )	0.07 $\pm$ 0.03*	0.12 $\pm$ 0.03 *
Mn (ppm)	60.54 $\pm$ 4.81 *	52.25 $\pm$ 6.06 *
Zn (ppm)	12.58 $\pm$ 2.00	12.21 $\pm$ 1.09
Cu (ppm)	2.46 $\pm$ 0.53	2.83 $\pm$ 1.22

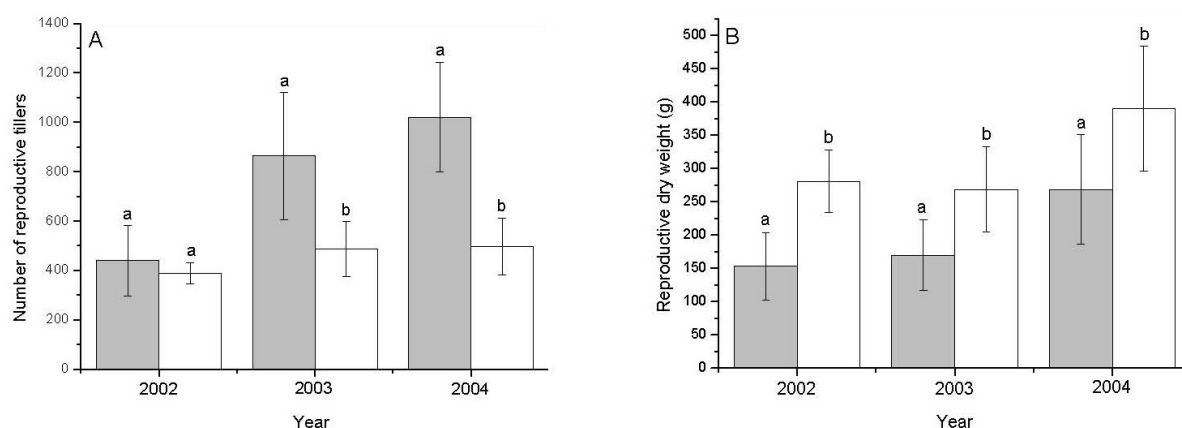


Figure 1. Growth parameters of *Epichloë*-infected and healthy plants growing in the field over a three-year period. Means and standard deviations (bars) of (A) number of reproductive tillers produced, and (B) dry weight of infected and uninfected plants at the reproductive stage. Different letters indicate statistically significant differences among means.

## Conclusions

*Brachypodium phoenicoides* plants infected by *Epichloë typhina* remain asymptomatic during the vegetative phase of their life cycle. In spite of the lack of obvious symptoms at this growth stage, we detected differences in nutrient content among infected and uninfected plants. During the vegetative growth phase, when disease symptoms appear, the number of reproductive tillers was greatly increased in infected plants, but the total dry matter production decreased.

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## **Session 2**

### **Grassland as part of the food chain**





# Botanically diverse forage-based rations for cattle: implications for product composition, product quality and consumer health

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## Abstract

Forage is an important component of the sustainability of ruminant production in many parts of Europe and products from forage-fed animals are increasingly valued by the consumer. This paper considers the effects of forage, and botanically diverse forage in particular, on the fatty acid composition, shelf-life and sensory characteristics of bovine meat and milk. Emerging data indicate that milk and meat produced from botanically diverse pastures have higher concentrations of fatty acids and anti-oxidants which are considered to be of benefit to human health. The data on differences in sensory characteristics of bovine products from botanically diverse pasture compared to intensive pasture *per se* are not convincing. However, differences in the production system, encompassing differences in environment/region and possibly animal type as well as the botanical diversity of pasture, can influence the sensory characteristics of cheese in particular. Among the challenges to capturing the benefits of botanically diverse pastures is the requirement to authenticate food products from particular environments/production systems and the long term maintenance of the botanical diversity in a pasture which contributes the unique set of compositional/sensory characteristics to the products of this system.

Keywords: Forage, botanically diverse, fatty acids, sensory

## Introduction

Forage, *in situ* or conserved, is a major component of ruminant production in most temperate regions. Grazed pasture is usually a cost effective feed option for producers and, in recent years, pasture-based systems have become to be regarded as more environmentally and animal-welfare friendly alternatives to intensive/feedlot systems of production. There is also growing interest by consumers in the safety, healthiness and quality of their food, its origin and in the methods by which it is produced.

The composition of a ration consumed by a ruminant can have a marked influence on the composition, appearance and sensory characteristics of milk and meat and their products. The influence of forage feeding *per se* on milk and meat with respect to fatty acid composition, processing characteristics and sensory quality has been regularly reviewed (e.g. Coulon and Priolo, 2002; Scollan *et al.*, 2005, 2006a; Elgersma *et al.*, 2006). Much of the literature on the quality of ruminant products from pasture-based systems of production relates to improved pastures, often of one dominant ryegrass species. This reflects the drive for increased productivity and efficiency and the uptake of cultivars bred for particular traits of economic importance. While reference will be made in this paper to such literature, emphasis will be placed where possible on the differences between pasture species and on the quality of products from pastures with a more botanically diverse composition.

### Fatty acid composition of milk and meat from cattle fed (botanically diverse) forage

The World Health Organization (WHO, 2003) recommends that total fat, saturated fatty acids (SFA), omega-6 polyunsaturated fatty acids (PUFA), omega-3 PUFA and *trans* fatty acids should contribute < 15-30%, < 10%, < 5-8%, < 1-2% and < 1% of total energy intake by man, respectively. Reducing the intake of SFA and increasing the intake of omega-3 PUFA are particularly encouraged. Meat, fish, fish oils and eggs are important sources of omega-3 PUFA for man while beef and other ruminant products such as milk are dietary sources of conjugated linoleic acid (CLA) (Ritzenthaler *et al.*, 2001). The dominant CLA in beef is the *cis*-9, *trans*-11 isomer, which has been identified as processing a range of health-promoting biological properties including antitumoral and anticarcinogenic activities (De la Torre *et al.*, 2006).

Feeding fresh grass or grass silage (rich in linolenic acid) compared to concentrates (rich in linoleic acid) results in higher concentrations of omega-3 PUFA and CLA in ruminant milk and meat lipids (French *et al.*, 2000a; Elgersma *et al.*, 2006). French *et al.* (2000a) also found significant reductions in the proportion of SFA with grass feeding. Collectively these responses in both SFA and omega-3 PUFA contribute towards beneficial changes in PUFA:SFA (increasing) and omega-6: omega-3 PUFA ratios (decreasing). In addition, feeding mixtures of grass and clover silage (both white and red clover) relative to grass silage alone increased the deposition of both omega-6 and omega-3 PUFA in muscle of beef steers and in milk fat (Dewhurst *et al.*, 2006; Scollan *et al.*, 2006b).

The impact of grazing botanically diverse pastures on milk fatty acid composition is illustrated in Table 1. Only studies with a control diet of 600 g kg<sup>-1</sup> dry matter (DM) or more of grasses (*Poaceae* family) and a botanically diverse forage with 300 g kg<sup>-1</sup> DM or more of herbs, and studies that individually reported milk fatty acids of particular interest (i.e. C18:3n-3, CLA *cis*-9, *trans*-11 and C18:1 *trans*-11) were considered. In these studies, lowland vs. highland pastures (Collomb *et al.*, 2002), ryegrass hay vs. natural grassland hay (Ferlay *et al.*, 2006), organically grown pastures vs. Swiss alpine pastures and l'Etivaz pastures (Kraft *et al.*, 2003), barn and pasture feeding in lowlands vs. alps (Leiber *et al.*, 2005) and intensively managed ryegrass silage vs. 60% of species-rich grassland silage (Lourenço *et al.*, 2005b) were compared. Overall, dietary treatments with a higher botanical diversity increased milk C18:3n-3 and PUFA concentrations whereas milk SFA concentrations were in most cases decreased. No consistent response in milk CLA *cis*-9, *trans*-11 and C18:1 *trans*-10+*trans*-11 was observed, with about half of the more diverse dietary treatments causing a reduction (Leiber *et al.*, 2005; Ferlay *et al.*, 2006), whereas considerable increases were reported in the other half of the dietary treatments (Collomb *et al.*, 2002; Kraft *et al.*, 2003; Lourenço *et al.*, 2005b). Differences between studies might be related to the relatively high concentrations of these fatty acids in the milk fat of the control groups. Indeed, milk CLA *cis*-9, *trans*-11 when feeding conserved ryegrass amounted to 0.87 and 1.36 g/100 g milk fatty acids in the studies of Ferlay *et al.* (2006) and Leiber *et al.* (2005), respectively vs. 0.26 g/100 g milk fatty acids in the study of Lourenço *et al.* (2005b). Also under ryegrass grazing conditions, Leiber *et al.* (2005) reported high CLA *cis*-9, *trans*-11 in milk fat (1.71 g/100 g milk fatty acids) compared to the other grazing studies (0.81 and 0.87 g/100 g milk fatty acids in the studies of Collomb *et al.*, (2002) and Kraft *et al.* (2003), respectively). Additional information on the fatty acid composition of milk from cows grazing alpine pastures can be found in Dewhurst *et al.* (2006).

Table 1. Fatty acid composition of milk and meat from ruminants consuming botanically diverse pasture. Data are expressed relative to intensively managed ryegrass pasture, shown in parentheses (g/100 g fatty acids, \* indicates a significant ( $P < 0.05$ ) change) and are adapted from Lourenco, *et al.* (2007c) and calculated from original papers.

Ref. Botanically diverse pasture	C18:3n-3	CLA	SFA	MUFA	PUFA
Bovine Milk					
1. Highland pasture	1.46(0.79)*	2.69(0.81)*	0.88(54.5)*	1.16(20.9)*	1.70(2.7)*
2. Natural grassland	1.23(1.02)*	0.82(0.87)*	1.01(66.2)	1.01(17.4)	1.05(2.9)
3. Swiss alpine pastures	1.36(0.86)	2.63(0.87)*	0.90(55.3)*	1.17(22.4)*	1.43(3.3)*
4. Swiss alpine pastures	1.51(0.86)	3.06(0.87)*	0.91(55.3)*	1.06(22.4)	1.62(3.3)*
5. Alpine pastures	1.53(0.67)*	0.85(1.37)	1.08(65.8)*	1.21(19.7)*	1.25(2.8)*
6. Alpine pastures	1.63(0.70)*	0.79(1.71)*	0.91(61.9)*	1.22(22.3)*	1.21(3.4)*
7. Species-rich grassland	0.96(0.61)	1.69(0.26)*	0.97(63.4)*	1.07(21.0)*	1.18(1.7)*
Lamb					
8. Saltmarsh pasture	1.11(1.42)	1.08(1.01)	1.02(41.9)	0.98(40.7)	1.11(6.5)
9. Botanically diverse silage	0.94(1.62)	1.98(0.30)*	0.95(46.1)	0.93(36.4)	1.30(10.6)
10. Botanically diverse pasture	1.02(2.59)	0.99(0.90)	0.88(43.0)*	0.81(35.6)*	1.82(11.9)*
11. Mountain pasture	-	-	1.01(45.3)	0.95(40.3)*	1.57(4.4)*

1. Collomb *et al.* (2002a), 2. Ferlay *et al.* (2006), 3. Kraft *et al.* (2003), 4. Kraft *et al.* (2003), 5. Leiber *et al.* (2005), 6. Leiber *et al.* (2005), 7. Lourenco *et al.* (2005a), 8. Whittington *et al.* (2006), 9. Lourenco *et al.* (2007a), 10. Lourenco *et al.* (2007b), 11. Adnøy *et al.* (2005).

There is a paucity of information on the effect of grazing botanically diverse pastures on the fatty acid composition of beef. Fraser *et al.* (2007) recently reported that inclusion of a period of grazing a *Molinia caerulea*-dominated semi-natural pasture increased the proportion of omega-3 PUFA and CLA in muscle lipids. The potential impact of grazing botanically diverse pastures on meat fatty acids is illustrated in Table 1, with data on lamb. These studies compared grazing of a perennial ryegrass (*Lolium perenne*) pasture with unimproved saltmarsh pasture (Whittington *et al.*, 2006) or a botanically diverse pasture (Lourenço *et al.*, 2007b), or lowland vs. mountain pastures on the West coast of Norway (Ådnøy *et al.*, 2005). Further, stable feeding of at least 70% ryegrass silage was compared with botanically diverse silage from a natural, unfertilized grassland (Lourenço *et al.*, 2007a). A general tendency for an increase in omega-3 and total PUFA proportions in intramuscular fat is observed mainly due to the higher proportions of C20 and C22 PUFA. For a comprehensive review of this topic the reader is referred to Lourenco *et al.* (2007c).

The mechanisms underpinning these effects on the fatty acid composition of milk and meat from ruminants consuming botanically diverse forage have not been completely elucidated. They may relate to the fatty acid composition of individual plants within the swards, although in none of the reported studies did botanically diverse forage supply more omega-3 PUFA than the more intensively managed forage (Lourenço *et al.*, 2007c). Nevertheless, the presence of leguminous plants has been associated with increased rumen PUFA outflow rates (Dewhurst *et al.*, 2006). For red clover, this might be associated with the presence of polyphenol oxidase, which reduces lipolysis during ensiling (Lourenço *et al.*, 2005a) and rumen fatty acid metabolism (Lee *et al.*, 2007). Polyphenol oxidase activity might also be present in other species of botanically diverse pasture products, but no information is yet available. There is some evidence that plant extracts (Sharma *et al.*, 2005) and specific secondary plant metabolites, such as condensed tannins or saponins (Han *et al.*, 2001; Moreno *et al.*, 2003; Ikeda *et al.*, 2005) inhibit lipase activity. However, the latter studies have been

performed with extracts from grape seeds or tea and the presence of specific metabolites in herbs of botanically diverse forage which might modify rumen fatty acid metabolism still needs to be examined. Besides effects on pre-ruminal (e.g. during wilting or ensiling) or rumen fatty acid metabolism, Lourenço *et al.* (2005b) suggested that feeding botanically diverse silages was associated with a higher transfer efficiency of C18:3 n-3 from the duodenum to the mammary gland compared to feeding silage from intensively managed grassland (104 vs. 69.6 %). Finally, interpretation of the origin of higher intramuscular PUFA concentrations should be considered with care, due to varying intramuscular fat content of animals fed botanically diverse pasture vs. intensively managed pasture products which can influence the PUFA concentration *per se*.

### **Flavour and shelf-life of milk and cheese from cattle fed (botanically diverse) forage**

The sensory quality of food can be defined by the texture, evaluated mainly in the mouth, the flavour, including the odour (smell attributes) and the aroma (sensations perceived by the retro-nasal airway) and the taste, perceived in particular on the tongue. The sensory quality of dairy products is influenced by the manufacturing process applied but it can also be strongly modified by animal diet (reviewed by Coulon *et al.*, 2004; Martin *et al.*, 2005a). The influence of feed type (e.g. maize silage vs. grass silage or hay) and of grass preservation method (green pasture, silage or hay) is now well documented for butter and cheese. Some of these feed factors have also been shown to impact the sensory properties of milk (Dubroeuq *et al.*, 2002). Apart from the well-known effect of certain specific plants like cabbage, garlic or onion on milk or cheese off-flavours (Urbach, 1990), the specific effect of the floristic composition of the grass has been relatively unstudied despite the fact that cheese makers believe that this factor has an important influence on cheese sensory properties.

The fatty acid composition of milk may play a role in flavour, e.g. oxidized milk and milk products are characterized by metallic, cardboard or stale flavours, and production of oxidized flavour at 8-days post sample collection was positively correlated with levels of C18:2n-6 ( $r = 0.49$ ), C18:3n-3 ( $r = 0.55$ ) and total PUFA ( $r = 0.50$ ) in milk fat (Timmons *et al.*, 2001). An effect of forage botanical composition on the sensory characteristics of cheeses may also involve non-lipid components derived from forages such as terpenes. These plant-specific molecules have aromatic properties and abound in certain aromatic dicotyledon species found in diversified meadows (Mariaca *et al.*, 1997; Cornu *et al.*, 2001). These molecules pass rapidly into milk (Viallon *et al.*, 2000) and are found in higher concentrations in cheese when the animals are fed dicotyledon-rich natural grass forage (Moio *et al.*, 1996; Viallon *et al.*, 1999). However, it appears that the increase in terpene concentration in cheese is not sufficiently large to exert any marked effect on flavour (Moio *et al.*, 1996, Bugaud *et al.*, 2001). In addition, because cheeses which were richest in terpenes had a globally milder flavour (less animal, spicy, cabbage, toasted, fermented vegetable, pungent and more nutty) and had a lower concentration of volatile products of protein breakdown by microbial enzymes, it was suggested that terpenes could have an antimicrobial effect. This may have an indirect impact on cheese sensory properties by modifying the dynamics of the microbial ecosystem during cheese making and ripening. Nevertheless, in a recent trial in which terpenes were added to milk prior to cheese making, it was not possible to confirm this hypothesis (Tornambe *et al.*, 2007a). In the case of a Mediterranean pasture particularly rich in aromatic plants, Carpino *et al.* (2004) recently reported that, in addition to the terpenes, other odour-active compounds found in pasture plants can be transferred to cheese (aldehydes, esters, sulphur compound). Plasmin is an endogenous milk protease that plays an important role in the ripening of cooked-curd type cheese. Its higher activity in milk of cattle consuming



plant species such as buttercup, present in certain types of meadows may contribute to differences in flavour characteristics of cheese (Buchin *et al.*, 1999; Bugaud *et al.*, 2001).

The influence of botanically diverse pasture on milk sensory properties has been studied in two recent experiments in France. A trained sensory panel, scoring 16 attributes, did not detect any differences in milk produced on commercial farms from pasture with a range in species biodiversity (Guichand *et al.*, 2006). In a controlled experiment, the sensory properties of milk from cows grazing 3 different mountain grasslands characterized by their increasing floristic diversity: a temporary grassland (17 species), a permanent grassland (31 species including 4 aromatic species) and a diversified permanent grassland particularly rich in aromatic species (50 species including 9 aromatic species) were compared (Tornambe *et al.*, 2007b). The sensory panellists were not able to discriminate between the milk from these three grasslands. The authors conclude that in wet mountain conditions, the variability of the floristic composition of the grassland does not impact the sensory properties of milk. It would be of interest to verify those first results in other conditions, in particular when aromatic plants are more important in the pasture as in some highlands or Mediterranean pastures.

Several trials have been conducted in Europe to examine the effect of the botanical diversity of forages (either grazed or preserved) on the sensory characteristics of various types of cheeses. Both holistic approaches which examined the links between the region, farming practices and cheese sensory properties and conventional experimental approaches focusing on the pasture characteristics under controlled conditions for milk production and cheese making have been undertaken. In the Reblochon region of France, Martin and Coulon (1995) found that differences in forage botanical composition could be associated with differences in cheese sensory characteristics under certain manufacturing conditions and so contribute to the protected designation of origin or PDO status of cheese from this region. Likewise, for Comté cheese, an important PDO cheese in France, Monnet *et al.* (2000) found evidence of associations between the botanical typology of pastures and the sensory typology of cheeses. Bérodiér (1997) showed that botanical diversity could be associated with more diversified and numerous cheese aroma attributes.

To our knowledge, direct experimental comparisons of cheeses made with animals fed monospecific vs. diversified grass do not exist. All the trials described in the literature were carried out with commercial farmhouse cheese makers in which the characteristics of cheeses made when the herd successively grazed pastures with different botanical compositions were compared (Martin *et al.*, 2005b). All the trials which involved different cooked pressed cheeses – Etivaz in Switzerland (Bosset *et al.*, 1999), Beaufort and Abondance in France (Buchin *et al.*, 1999; Bugaud *et al.*, 2001; Martin *et al.*, 2005a) – show evidence of modifications of texture and/or flavour accompanying pasture changes. In the study of Bugaud *et al.* (2001) it was possible to detect some associations between the botanical composition of the meadows and sensory features of the cheeses (Figure 1). In particular, the most gramineae-rich pastures were associated with the most intense 'cooked cabbage', 'pungent' or 'butyric' odours. The texture of these cheeses was also more elastic, firm and cohesive. The meadows rich in dicotyledon plants from a dry environment and high altitude were associated with cheeses with a more fruity and toasted flavour, whereas the pastures rich in dicotyledon plants from a wet environment and/or frequently unconsumed were associated with cheeses with bitter and animal flavours and a melting texture. Nevertheless, at present, generalization of these results remain uncertain; even within the case of the Abondance cheese, a recent meta-analysis of the results of 4 similar trials (50 cheeses and associated pastures) did not permit a generalization to be made on the relationship between the botanical diversity of pasture and

the sensory characteristics of cheese (Farruggia *et al.*, unpublished data). In the case of preserved forages, Verdier-Metz *et al.* (2000) made uncooked-pressed cheeses according to a Saint-Nectaire technology in an experimental farm with milk from cows managed under similar conditions (milking, health status, nutrient inputs, housing) but fed cocksfoot hay or natural Auvergne grass (green or hay). Compared to cocksfoot-hay cheeses, cheeses from natural grass were less melting and less bitter, with less developed rancid or mouldy odours. However, the differences were lower than those observed in studies at pasture with cooked cheeses.

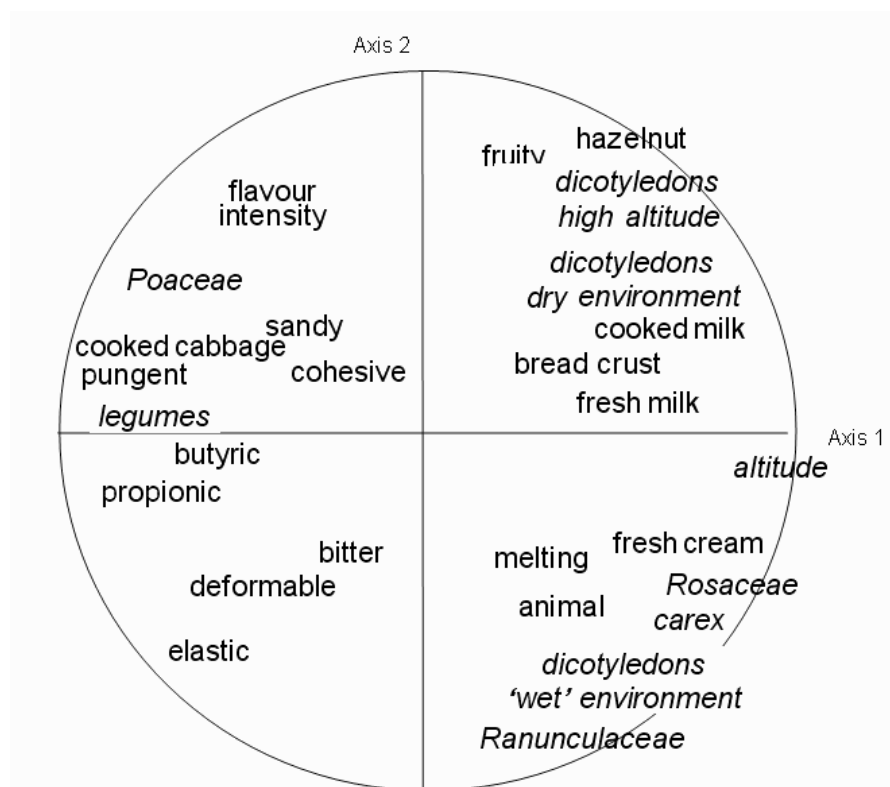


Figure 1. Principal component analysis of the correlations between the abundance of plant families in pastures and the texture and flavour of the corresponding Abundance cheeses (from Martin *et al.*, 2005). The botanical compositions of the pastures were the active variables and the sensory properties of cheeses were added as additional variables (Axis 1 and 2 represent 37 and 18 % of the variability respectively).

Determining the relationships between pasture characteristics and the sensory properties of dairy products is of special importance in the case of products with a PDO or a Protected Geographical Indication [PGI] label, which claim close links with milk production conditions. Among those production conditions, forage botanical composition is part of the basic link between products and their original 'terroir'.

The fatty acid composition of milk can also influence its processing characteristics. An increase in oleic acid concentration generally results in 'softer' or more spreadable butter with a more desirable melting profile, whereas milk having a high PUFA concentration is more susceptible to oxidation than conventional milk (Murphy, 2000). Milk from cows fed on red clover compared to grass silage contained more C18:2n-6 and C18:3n-3 PUFA which also resulted in increased oxidative deterioration of milk (Al-Mabuk *et al.*, 2004). The data in Table 1a would suggest therefore that milk from cows fed botanically diverse pasture would be more oxidatively unstable than milk from intensive pasture. While Havemose *et al.* (2004)

observed that milk from cows fed grass silage had higher concentrations of the antioxidants  $\beta$ -carotene, lutein, zeaxanthin and  $\alpha$ -tocopherol, than cows fed maize silage, the levels were not sufficient to prevent higher lipid oxidation in the former. The concentration and spectrum of antioxidants in milk from cows fed botanically diverse pastures is unknown. However, antioxidant properties have been reported for many varieties of herbs (Al-Mamun *et al.*, 2007; Xu and Chang, 2007) which if present in botanically diverse pasture may confer added oxidative stability to milk. Further studies are needed in this respect.

### **Shelf-life and flavour of meat from ruminants fed (botanically diverse) forage**

The bright red colour of beef and lamb is important for consumers when making purchasing decisions. Priolo *et al.* (2001) concluded from their review of 35 studies that muscle from grazing cattle is darker. In contrast, Muir *et al.* (1998) considered 15 publications where beef from cattle fed forage-based diets was compared with beef from cattle fed cereal-based diets but, in which, the animals had been finished to comparable slaughter weights or fat cover levels. They concluded that there were no consistent effects of these feeds on meat colour. Where energy intake was similar and cattle were accustomed to 'handling' during grazing and so were not subjected to additional stress during loading and transport compared to their housed counterparts, muscle colour was similar (French *et al.*, 2000b). It is unlikely, therefore, that botanically diverse forage would affect muscle colour *per se*. Forage feeding leads to beef carcasses with more yellow fat than those from grain-fed animals, through the ingestion, absorption and deposition of carotenoids (mainly  $\beta$ -carotene and lutein). Feeding animals a low carotene-containing ration can reduce fat yellowness of previously forage-fed animals (Dunne *et al.*, 2006).

A consistent finding in the literature is that feeding green forages contributes antioxidants such as  $\alpha$ -tocopherol and  $\beta$ -carotene to the meat, which stabilize the fatty acids, increase shelf-life and make the meat more desirable when compared with concentrate feeding (O'Sullivan *et al.*, 2003; Gatellier *et al.*, 2005; Warren *et al.*, 2007). The type of conserved forage can also affect lipid and colour stability. Charolais-cross heifers were fed *ad libitum* on either maize silage, grass silage or a 50:50 mixture of these. Vitamin E concentration was 2.1, 3.8 and 3.0 mg kg<sup>-1</sup>, respectively. Meat from the animals fed the maize silage had the poorest lipid and colour stability, grass silage the best and the 50:50 mixture was intermediate (O'Sullivan *et al.*, 2002). Animals grazed on grass/clover mixed swards had increased proportions of omega-3 PUFA in their meat, but less vitamin E in their muscle, and this produced slightly less stable meat (Enser *et al.*, 2001; Scollan *et al.*, 2002). Animals fed red clover silage, grass silage, or a 50:50 mixture of the two, had an increasing content of PUFA in muscle with increasing red clover silage content but vitamin E concentration decreased as did colour and lipid oxidative stability. When a fourth group of animals was fed the red clover silage supplemented with vitamin E, vitamin E content of muscle, colour and lipid stability was the same as that in the muscle of 100% grass silage-fed animals (Richardson *et al.*, 2005).

With respect to botanically diverse pasture, muscle from traditional British beef breeds which grazed botanically diverse pastures growing on a carboniferous limestone site (400m) or two chalk downlands (130 and 180 m), an improved pasture (20m) or an unimproved pasture, all had good oxidative stability but the mean concentrations of vitamin E in muscle of cattle grazing the botanically diverse pastures were some of the highest measured to date (4.8-6.3 mg/kg, R.I. Richardson, unpublished observations). Inclusion of a period of grazing a *Molinia caerulea*-dominated semi-natural pasture increased the concentration of vitamin E in muscle of Welsh Black cattle which was reflected in lower fat oxidation. In all cases there was at least 8 d of colour shelf life, which is longer than that required by major retailers (Fraser *et*



*al.*, 2007). Finishing lambs for 3 months on pastures that differed in botanical composition did not result in differences in colour stability or lipid oxidation but protein oxidation was higher in meat for the lambs grazing the botanically diverse pasture (Petron *et al.*, 2007). In a study of sheep reared on different pasture types, the 3 groups fed on species-rich pastures (saltmarsh, heather and moorland) had muscle vitamin E values greater than 4.8 mg kg<sup>-1</sup>. The heather group had a mean value of 7.0 mg kg<sup>-1</sup>, the highest mean value observed compared to other published studies. Analysis of lipid oxidation products revealed there was no significant difference between the four pasture groups and all groups were below 2, the level at which lamb meat is considered to be approaching incipient rancidity (Whittington *et al.*, 2006).

Tenderness is the most important sensory influence on the acceptability of meat, but when tenderness is increased, flavour and juiciness increase in relative importance. When possible confounding influences are removed, such as comparing animals at similar weights or fat cover, there is little evidence for a consistent difference in tenderness or juiciness between grass-fed and grain-fed beef (Muir *et al.*, 1998; French *et al.*, 2000b; Geay *et al.*, 2001).

The flavour of red meats which develops during cooking, derives from the Maillard reaction between amino acids and reducing sugars and the thermal degradation of lipid. The former produces roasted/meaty flavours and the latter the species differences in flavour (Mottram, 1998). Diets which alter the fatty acid composition of the lipid fraction of meat could also alter the amount and type of volatiles produced and hence its aroma and flavour (Elmore *et al.*, 1999). In a study comparing grass- and concentrate-finished animals, the concentrate-fed animals had higher concentrations of linoleic acid in their meat and on cooking produced seven compounds at over three times the level found in meat from grass-fed animals, which had much higher concentrations of linolenic acid and produced a higher amount of only one compound, 1-phytene, a derivative of chlorophyll ingested with grass (Elmore *et al.*, 2004). Other compounds, not derived from fatty acids, may also contribute to flavour. Skatole (3-methyl indole) is commonly associated with boar taint in pigs. While it was noted that fat from grass-fed cattle had high concentrations of 3-methyl indole, the concentrations in the fat of silage-fed animals was more similar to that of concentrate fed animals (Whittington *et al.*, 2004). In the European Union many consumers feel that meat from less intensively-fed animals has a better taste, whilst in the US grass-finished beef is less acceptable (Melton, 1990). Raes *et al.* (2003) compared the fatty acid composition and flavour of retail beef from Limousin and Belgium Blue carcasses, which was produced locally in Belgium, with imported Argentinean and Irish beef. The fatty acid profiles suggested that the former meat was derived from cereal-fed animals and the latter were predominantly grass-fed. Sensory analysis and flavour volatile analysis showed that the grass-fed animals had higher flavour intensity with higher contents of low molecular weight unsaturated aldehydes derived from oxidation of long chain PUFA. In the study of Warren *et al.* (2008), loin joints from steers fed grass silage or fresh grass had higher scores for beef flavour and lower scores for abnormal flavour when compared to concentrate diets (restricted to the growth rate of the silage-fed animals) (Richardson *et al.*, 2004). They also scored higher for 'overall liking' than the concentrate group. Although not statistically significant, the higher values for 'livery' seen in the grass groups are consistent with other unpublished results.

When forages were compared, Scollan *et al.* (2002) found little difference in flavour of beef from cattle fed grass or mixtures of grass and red or white clover. Similarly, in the study of Fraser *et al.* (2007) mentioned earlier, the taste panel failed to detect major differences between meat from the *Molinia caerulea*-dominated semi-natural pasture and that from animals grazing improved pasture. In a US study, steers finished on white clover had a higher

'grassy' flavour than those fed grass, and grass-fed animals produced meat which was not only higher in PUFA but was also oxidatively more unstable (Larick and Turner, 1990). The authors attributed these differences in flavour to both the increased content of PUFA, particularly  $\alpha$ -linolenic acid, its lower oxidative stability, and to odoriferous compounds stored in the depot fats.

The effect of type of forage on flavour is better documented for sheep (see Young *et al.*, 2003). Diet appears to have a greater effect than breed and differences in flavour, explained by higher concentration of C18:3 in animals fed grass and C18:2 in animals fed concentrates, appear to be more intensive in lamb. Whilst the odour/flavour of lamb is due to a wide range of compounds, the characteristic species flavour is caused by branched-chain fatty acids. Cross-bred Suffolks finished on lowland grass or concentrates were compared with Soay, finished on lowland grass, and Welsh Mountain finished on upland flora (Fisher *et al.*, 2000). Flavour characteristics were similar for lamb chops from Welsh Mountain and Suffolks fed grass, which differed from Soay and Suffolks fed concentrates. The latter had low scores for flavour and high scores for abnormal flavour, metallic, bitter, stale and rancid. Lambs grazed on white clover or alfalfa had more intense flavour than those grazed grass pastures, whilst lambs finished on grass pasture at a slow rate of growth had more intense flavour than those finished on pasture at a high rate of growth or on concentrates (see Roussett-Akrim *et al.*, 1997; Duckett and Kuber, 2001). In the study of Whittington *et al.* (2006) involving lambs grazing 3 different species-rich pastures (saltmarsh, heather and moorland) the taste panel scored all pastures high in lamb flavour and scored the control lamb (predominantly ryegrass) significantly lower. In addition, the control lamb scored higher for abnormal odour of the fat, and higher for abnormal flavour of the lean with more rancid notes. Adnøy *et al.* (2004) compared the quality of meat from lambs grazed on unimproved mountain pasture with meat from those on cultivated lowland grassland and found significant differences in carcass fat content and fatty acid composition, meat colour and flavour. They suggested that meat from lambs raised in extensive systems on mountain ranges may have certain qualities that might be used in the promotion of local or regional products. Young *et al.* (1994) reported that feeding lambs on a range of different pasture species resulted in different sensory attributes of meat.

### **Authentication of ruminant derived products of botanically diverse pastures**

To protect a product brand and to maintain the confidence of the customer, direct methods are needed to authenticate the provenance of food. This is particularly important when food products are marketed on the basis of their region of production such as a specific PDO or PGI label. Some constituents of forage, and in particular botanically diverse forage, are reflected in the milk and meat produced on those forages as illustrated earlier for fatty acids, terpenes and carotenoids. These molecules are potential 'biomarkers' of the dietary history of the animals (Prache, 2007). Issues such as the time of appearance of a marker relative to a change in diet, persistence, presence in different tissues, discriminatory ability and development of rapid analytical producers are under active investigation for terpenes and carotenoids in particular. Stable isotope ratio analysis (SIRA) also provides a promising tool for authenticating the dietary history of food. SIRA exploits variations in the natural stable isotope composition in farm products, which can reflect different crop cultivation and animal husbandry practices. For example, SIRA of C can potentially identify the production origin of beef and milk based on the extent of C<sub>4</sub> photosynthetic plant materials (maize) consumed by cattle (Bahar *et al.*, 2005; Knobbe *et al.*, 2006). Other approaches under investigation include spectroscopic techniques, volatile components of tissue, DNA-based technology and alkane concentrates in adipose tissue (De la Fuente and Juarez, 2005; Reid *et al.*, 2006). When

coupled with chemometric analysis, the above methods alone or combined should yield robust tools for the authentication of products of botanically diverse pastures, in the near future.

## Conclusions

Modern consumers have a greater interest in the environment, animal welfare and the origin and method of production of their food than heretofore. This is reflected in growing preference for food products of pasture-based systems of production and of regional/quality based systems of production such as those labelled as Protected Denominations of Origin (PDO) or Protected Geographic Indications (PGI). Botanically diverse or unimproved pastures are central to many of these production systems. While a substantial body of information is available on the differences in composition and sensory properties of products from pasture-based and concentrate-based systems of production, relatively little information is available on the differences in product quality between different botanically diverse pastures or indeed between botanically diverse pastures and improved or more productive selected monospecific pastures. Nevertheless, emerging data indicate that milk and meat produced from botanically diverse pastures have higher concentrations of fatty acids and antioxidants which are considered to be of benefit to human health. This should not only enhance still further the attractiveness of these products for the consumer, but also provide an opportunity to develop a range of novel functional foods. The term functional food is a generic term used to describe foods or food components that have beneficial effects on human health above that expected on the basis of nutritive value (Milner, 1999). Such products are targeted at disease prevention and are aimed at healthy people. Milk and meat and associated products particularly from botanically diverse pastures offer exciting opportunities in this rapidly developing area. The data on differences in sensory characteristics of bovine products from botanically diverse compared to intensive pasture *per se* are not convincing. However, differences in the production system, encompassing differences in environment/region, possibly animal type as well as the botanical diversity of pasture, can influence the sensory characteristics of cheese in particular; the 'terroir' effect. Development of analogous systems to create product differentiation would seem to be a useful strategy for the meat industry.

Among the challenges in this regard is the requirement to authenticate food products from particular environments/production systems and the long term maintenance of the botanical diversity in a pasture which contributes to the unique set of compositional/sensory characteristics to the products of this system. This is essential to the sustainability of markets for these products and to the further development of PDO/PDI marketing initiatives. As information accumulates on the composition and impact of individual herbs and novel plant species on milk and meat quality, opportunities will arise to develop novel bio-diverse pastures for particular product quality characteristics.

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# Plant diversity in grasslands and feed quality

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## Abstract

Plant diversity in complex grassland swards is structured in three levels of organization, i.e. plant functional groups, species within functional group and genotypes within each species. This structure of diversity has to be considered when analysing feed quality for the ruminants and trying to improve it.

The paper will first review the relationship between species diversity, forage composition and feed value for the ruminant, both in permanent and temporary grasslands. Special attention will be drawn on the effect of specific composition, plant functional groups and agronomic practices. Nutrient use and interactions of major nutrients with secondary plant metabolites such as condensed tannins or phenol oxidase will be considered.

In the second part, perspectives of forage composition improvement in a context of multi-species temporary grasslands will be analysed with a special focus on water soluble carbohydrate fraction and polyphenol oxidase which may reduce proteolysis and lipolysis and thus improve feed value. The possible interactions between macro and micro nutrients are discussed as they are relevant in grasslands with complex species composition.

As a conclusion, main objectives for future research and breeding are identified and prospects offered by crop modelling are discussed.

Keywords: Species diversity, forage composition, macro-nutrients, micro-nutrients, breeding

## Introduction

Grasslands contribute a major part of the agricultural area in Europe (Huyghe *et al.*, 2006) and are the main source of feed for domestic herbivores. For this purpose, maximizing biomass production per unit area and optimizing feed quality to meet animal requirements have been major objectives over the last decades.

Alongside this production function, grasslands provide various ecosystem services among which the control of nitrogen losses and soil erosion, the reduction of energy consumption and greenhouse gas emissions, and preservation of biodiversity are particularly important (Huyghe *et al.*, 2005). Several studies have shown that ecosystem functions of grasslands will be better achieved by swards having a complex taxonomic composition compared to more simple swards. Diverse swards are likely to better persist (Alard and Balent, 2007). In addition, the fauna diversity is higher which is related to grazing management (Tallowin *et al.*, 2005), to the vertical structure of the vegetation (Rooney and Waller, 2003), to the spatial heterogeneity (Wallis de Vries, 1995) and the composition of the litter.

If grasslands of increased biodiversity are considered as having the potential to better serve production and ecosystems services than species poor grasslands do, there is the need for an improved understanding of the agronomy and utilization of such grasslands. The present contribution will focus on the feeding value of complex grasslands. In particular, the following aspects will be addressed. The first one is to identify the impact of sward diversity on the various components of feed value. The second is related to the possibility to achieve gain through plant breeding when varieties are to be grown in complex mixtures.

## Species diversity in grasslands and feed value

Analysing grassland biodiversity with regard to the agronomy of grassland systems requires a common understanding of biodiversity. Different organizational levels of diversity can be distinguished (Figure 1) each of which may affect the relationship of diversity and agronomy in a different way. Thus, we will consider these levels of diversity when analysing the feed quality for ruminants. In addition, we will analyse various components of feed value, i.e. the chemical composition of the herbage, the digestibility as a major indicator of the utilization of nutrients and the voluntary intake.

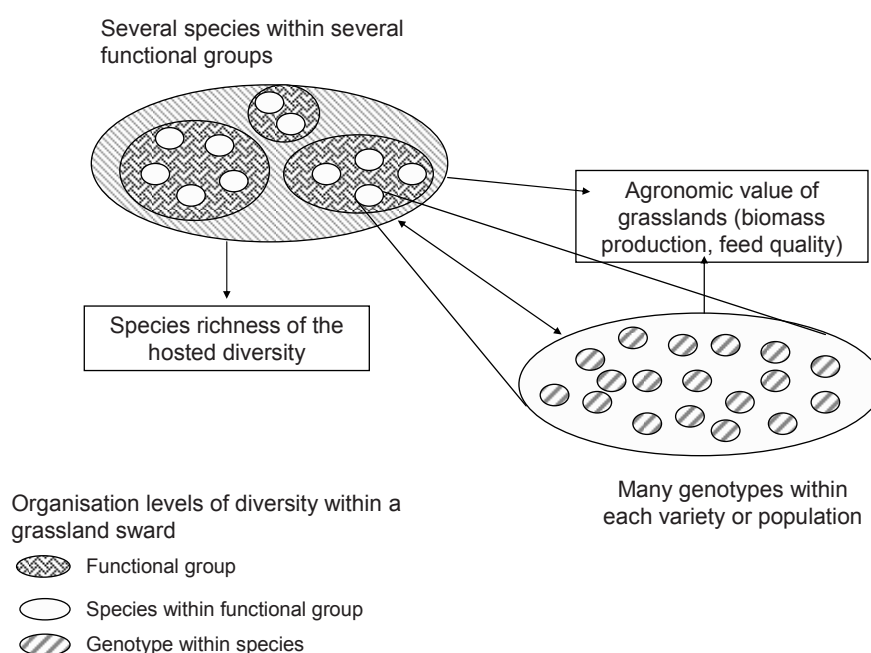


Figure 1. Organization of plant diversity in a grassland sward and impact on its agronomic and environmental values.

Sown and permanent grasslands are often regarded as very different, especially in terms of species diversity. This is obviously the case when temporary grasslands are sown with a single or few species, the diversity being then the result of recruiting species from either soil seed bank, from seed rain of adjacent vegetation or through zoochory from other grassland sites (Blanckenhagen and Poschold, 2005).

However, in most European countries, temporary grasslands tend to be increasingly sown with complex seed mixtures. It is thus of interest to assess their species diversity and the contribution of the diversity to biomass production.

Prior to the assessment of the impact of species diversity on feed value, it is necessary to analyse the possible impact of diversity on biomass production. This is of importance for feeding as it directly affects herbage availability. In addition, herbage mass is related to the sward structure and the physiology and morphology of the plants. This in turn affects the feeding value.

### *Relationship between species diversity and biomass production*

Many papers report a positive influence of the species richness on biomass production in grasslands. Prominent examples have been given by Hector *et al.* (1999) in eight European sites and by Guo *et al.* (2006) in four sites in North Dakota (USA) who analysed biomass

production of sown grasslands grown under mesotrophic conditions. They showed a trend of increased biomass production with an increasing number of species and plant functional groups including grasses, legumes and forbs. However, in most cases (years and locations), the maximum biomass was achieved from intermediate levels of diversity.

In 28 sites, across four European climatic regions, Kirwan *et al.* (2007) also observed a positive impact of the number of species and of functional groups (slowly and rapidly growing grasses and legumes), suggesting interactions of species. The positive agronomic interactions between grasses and legumes are obviously explained by the different ways of nitrogen nutrition and rhizodeposition (Soussana and Hartwig, 1996). Interactions of either grasses or legumes are readily less explained and deserve increased scientific attention. In perennial species, complementarity of resource use is the main mechanism by which the positive relationship between diversity and productivity is explained.

In grasslands sown with mixtures of 14 species from a species pool of adjacent native grasslands, Guo *et al.* (2006) assessed the contribution of every species to biomass production, and showed that the contribution of the species to biomass production was different in sown and permanent grasslands. The permanent swards were more stratified where one species had a higher contribution to herbage production. This means that the pattern of interaction between species may be different in sown and permanent grasslands. This may influence the chemical composition of the herbage.

#### *Impact of species richness on chemical composition of the herbage and animal performance*

The relationship between species diversity and chemical composition of herbage has generally received little attention in scientific research. However, if the potential of grassland biodiversity to benefit grassland farming is to be exploited, there is a strong need to investigate the feeding capacity of such grasslands. Three aspects should be considered: 1) is the relationship between biomass production and chemical composition affected by diversity in species and functional groups, 2) is the relationship between various chemical components affected by species diversity, 3) how constant is the chemical composition over seasons and years in swards with various species diversity?

This section is mainly based on results of on-going research at Lusignan, France, where plots with various initial species composition, with grasses and legumes, were grown under two fertilization levels and two cutting regimes. The number of sown species ranged from one to eight.

A negative relationship between biomass production and dry matter digestibility was found (Figure 2). No effect of the number of plant functional groups and of the species diversity within groups was found and both cutting regimes followed the same trend. A negative relationship was also observed between biomass production and protein content, but, as expected, with contrasting behaviours between pure grass swards and mixtures of grasses and legumes. The increase in biomass obtained due to species diversity may thus result in a slight decline in feed value. For the data of Figure 2, the highest values in biomass production and the lowest ones in digestibility were observed for swards with a high abundance of cocksfoot and tall fescue.

The relationship between the chemical components of the herbage was not affected by species or functional group diversity. The negative relationship between contents in protein and in water soluble carbohydrate only depended on the cutting frequency. This relationship held true for all functional groups. The effect of cutting frequency was likely to be related with the higher proportion of cell walls under less frequent cutting.

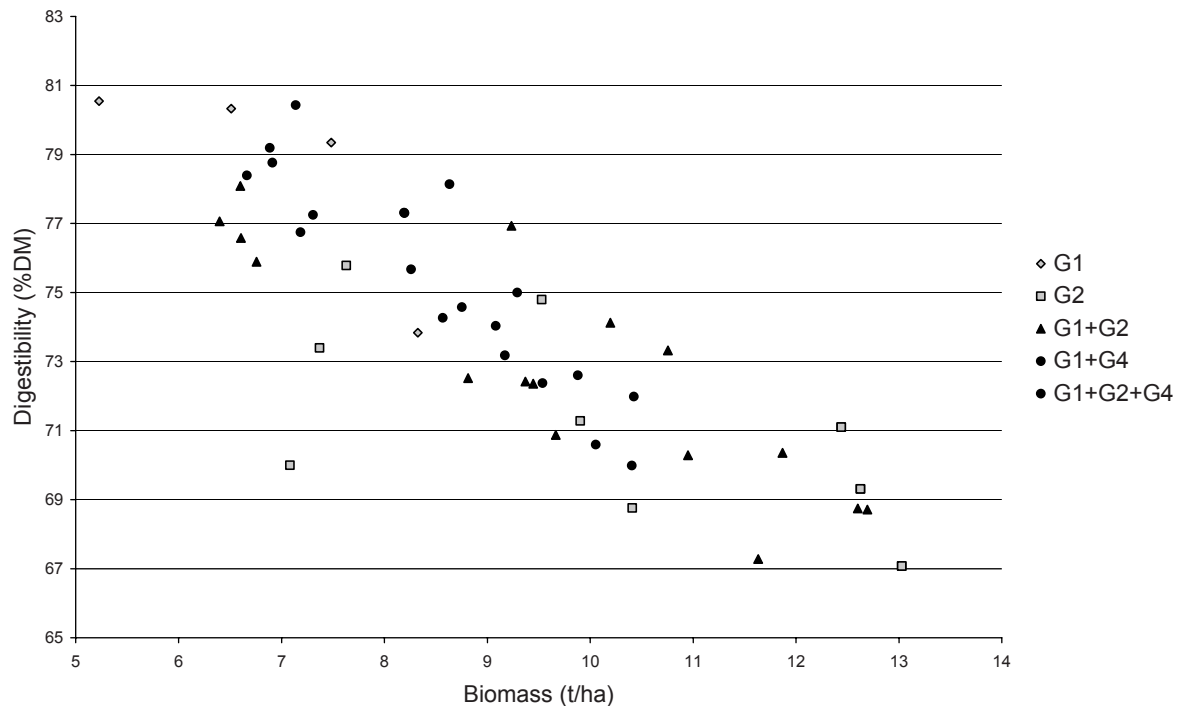


Figure 2. Relationships between annual biomass production and weighted digestibility in swards, for various compositions in functional groups under two cutting managements. G1: grasses with rapid growth (perennial ryegrass); G2: grasses with erect growth habit (cocksfoot, tall fescue, meadow fescue), G4: legumes with prostrate growth (white clover).

In addition, mixtures of grass and legumes had a lower variation of feeding value traits (digestibility, protein and water-soluble carbohydrate contents) between the different cuts compared to the respective monocultures of grasses. The same was found when more complex swards were investigated. For the grasses, species identity has a stronger effect on the variation of feeding value than the species or functional group number. This is the case when combining ryegrass whose chemical composition varied among cuts with cocksfoot which was more stable.

Finally, in swards sown with species adapted to intensive farming and under sufficient nitrogen supply, the variation of the chemical composition was mainly due to the variation in biomass production. Thus, the increased biomass found in more complex swards resulted in a slightly lower feeding value. However, the main advantage of complex swards combining grasses and legumes was the more stable chemical composition between cuts. Thus, legumes also have a stabilizing effect on chemical composition and feed value. It is also worth mentioning that in some tropical situations with diets rich in grasses with low nitrogen content, the addition of legumes with high N content induced a better digestibility of the grasses.

On sown grasslands grazed by dairy cattle, Soder *et al.* (2006) did not report any significant difference in voluntary intake between swards where the number of species ranged from two to nine. Milk production and composition were not affected. The only impact of the species diversity on animal production was detected for the conjugated linoleic acid content of milk fat which was higher for cows that grazed the multi-species swards compared to the cocksfoot/white clover mixture.

Species diversity may have a positive effect on voluntary intake as diversity offers a choice of herbage species and choice was shown to significantly increase intake at pasture (Cortes *et al.*, 2006). Bruinenberg *et al.* (2003) found significantly higher intake in cows for species-rich grasslands compared to species-poor ones, although digestibility and crude protein content were lower. Intake of digestible organic matter also increased. Ginane *et al.* (2002) with heifers and Champion *et al.* (2004) with sheep showed a relationship between species diversity of the diet and voluntary intake. However, it is not clear whether the variation in intake is a consequence of the diversity *per se* or of the choice situation, even though the experiments of Cortes *et al.* (2006) would support the hypothesis of a true effect of diversity. In contrast, Soder *et al.* (2006) found no effect on the intake of dairy cows. So, it may be hypothesized that the nutritional demand of the animals and their physiology could play a role in the relationship between species diversity of the sward and the intake. Andueza *et al.* (2005) showed that sheep voluntary intake in grasslands richer in forbs was higher than in grasslands with a high proportion of grasses. This shows that the role of species identity *per se*, such as forbs in the experiments of Andueza *et al.* (2005), could be as important as the role of diversity.

#### *Feed quality in permanent grasslands and relationship to the species diversity*

Species composition and diversity of permanent grasslands strongly vary in relation to the pedo-climatic conditions and the management practices. Thus variation in feed value of permanent grasslands can be expected to increase with species diversity because of differences in the feed value and in the phenology of the forage species (Bruinenberg *et al.*, 2002). Understanding feed value of permanent grasslands requires to analyse the following: i) differences in feed value of species which co-occur in grassland communities, i.e. native grasses, legumes and forbs, ii) characterization of feed value of grasslands from their botanical composition in relation to environmental and management factors, and iii) complementarities and interactions between species in diversified grasslands of particular interest with respect to feed value.

Bruinenberg *et al.* (2002) reviewed several studies in which *in-vitro* digestibility of grasses and forbs from permanent grasslands were measured. At a given date, differences in digestibility between species can reach 22%, as for example between *Nardus stricta* and *Lolium perenne*). Whereas *Lolium perenne* had the highest digestibility in most studies, digestibility of dicotyledonous species, for example *Ranunculus repens*, can reach similar values. More recently, the feed value of monocultures of thirteen perennial C3 grass species that co-occur in temperate semi-natural grasslands was compared in a factorial design of two levels of N supply and two cutting frequencies (Pontes *et al.*, 2007a). Species was the largest source of variation in pepsin-cellulase solubility of herbage, while crude protein concentration responded more to management factors. The highest values of digestibility were met for *Lolium perenne* and *Phleum pratense* and the lowest for *Festuca rubra* and *Poa pratensis*. Differences in digestibility were related to plant phenology, the highest digestibility values over the season being achieved for the late flowering species (Figure 3a). The digestibility of grasses decreases with time during the growing season (Baumont *et al.*, 2007), but the rate of decrease varies with the relative earliness of species. For example, while the digestibility values of *Alopecurus pratensis* and *Dactylis glomerata* are similar at a vegetative stage, four weeks after ear emergence of *Dactylis glomerata* the digestibility value of *Alopecurus pratensis* is 16% lower than that of *Dactylis* (Schubiger *et al.*, 2001). Compared to grasses, the decrease in digestibility is lower for legumes, especially for *Trifolium repens*, and the decrease of digestibility is very variable for forbs. The digestibility value of leafy forbs, like *Taraxacum officinale* are similar to the values achieved by vegetative grasses with almost no



decrease during the growing season (Schubiger *et al.*, 2001). In contrast, tall growing species of the *Apiaceae* family, and other species such as *Polygonum bistorta* that is common on alpine grassland, are characterized by a rapid decrease in digestibility (Daccord *et al.*, 2006).

Analysis of plant functional traits helps in understanding species differences in feed value. Leaf traits such as specific leaf area and leaf dry matter content can be used to classify grasses co-occurring in grasslands into functional types that correspond to plant growth strategies according fertility level and defoliation rate (Cruz *et al.*, 2002; Louault *et al.*, 2005). Species adapted to fertile soils and frequent defoliation (e.g. *Lolium perenne*, *Poa trivialis*) are characterized by a high specific leaf area and a low leaf dry matter content that favour a fast regrowth after defoliation. In contrast, species adapted to infertile soils and infrequent defoliation (e.g. *Brachypodium pinnatum*) show low specific leaf area and high leaf dry matter content. This classification is associated with a gradient of digestibility values as digestibility increases with specific leaf area and declines with leaf dry matter content (Pontes *et al.*, 2007b, Figure 3b).

From the functional approach, it can be expected that the feed value of permanent grasslands will largely depend on their composition in functional types influenced by environmental and management factors. In a long term experiment, Louault *et al.* (2005) showed that reducing fertilization and grassland utilization will select functional types with lower specific leaf area, higher leaf dry matter content and lower digestibility.

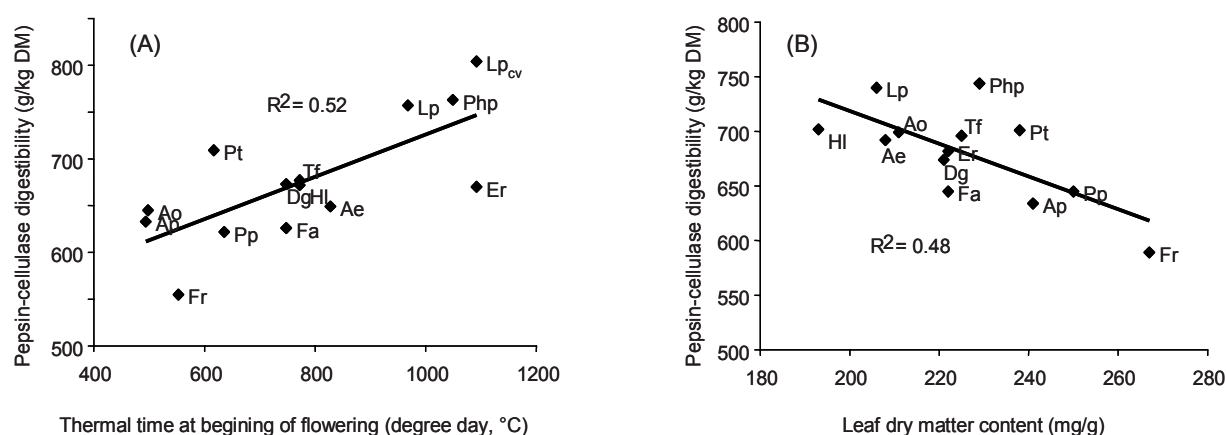


Figure 3. Relationship between pepsin-cellulase digestibility and thermal time at flowering (A) and leaf dry matter content (B) for 13 native grass species of semi-natural grasslands. *Alopecurus pratensis* (Ap), *Anthoxanthum odoratum* (Ao), *Arrhenatherum elatius* (Ae), *Dactylis glomerata* (Dg), *Elytrigia repens* (Er), *Festuca arundinacea* (Fa), *Festuca rubra* (Fr), *Holcus lanatus* (Hl), *Lolium perenne* (Lp), *Lolium perenne* cultivar (Lp<sub>cv</sub>), *Phleum pratense* (Php), *Poa pratensis* (Pp), *Poa trivialis* (Pt) and *Trisetum flavescens* (Tf). Adapted from Pontes *et al.* (2007a and 2007b).

For medium to intensively used grasslands in Switzerland, Daccord *et al.* (2006) proposed a simple and useful typology that characterizes the feed value of grassland in relation to their botanical characteristics. Four main groups were defined from the proportion of grasses, legumes and forbs. In addition, for the two groups dominated by grasses (> 70% and between 50 and 70%), sub-groups differentiated grasslands in which *Lolium perenne* represented more than 50% of grasses as ryegrass will contribute to higher feed value, and for the group dominated by forbs (> 50%), sub-groups differentiated leafy forbs from forbs with strong stems. Chemical composition and feed value, and their change with maturity stage (Figure 4),

were then calculated from individual plant values for virtual mixtures representing the different types of grasslands.

The ability of diversified grasslands extensively managed for biodiversity purposes to ensure sufficient feed value for animal production has been investigated in several studies (Tallowin and Jefferson, 1999; Bruinenberg *et al.*, 2003; Fiems *et al.*, 2004). In most studies, nutritive value of extensively managed grasslands was reduced. But these effects may be more due to reduced fertilization and lower defoliation frequency than to increased species diversity. The study by Daccord *et al.* (2006) rather suggested a positive role of diversity in grassland on feed value. Digestibility (Figure 4), energy and protein values are likely to be higher and less prone to decrease with maturity stage in particular when leafy forbs play an important role in the swards. A lower variability of the digestibility in grasslands dominated by species adapted to low fertility soils was also hypothesized by Duru (1997). For late heading species in particular, low growth rates will maintain a high leaf to stem (or leaf to sheath) ratio for a longer period during the season.

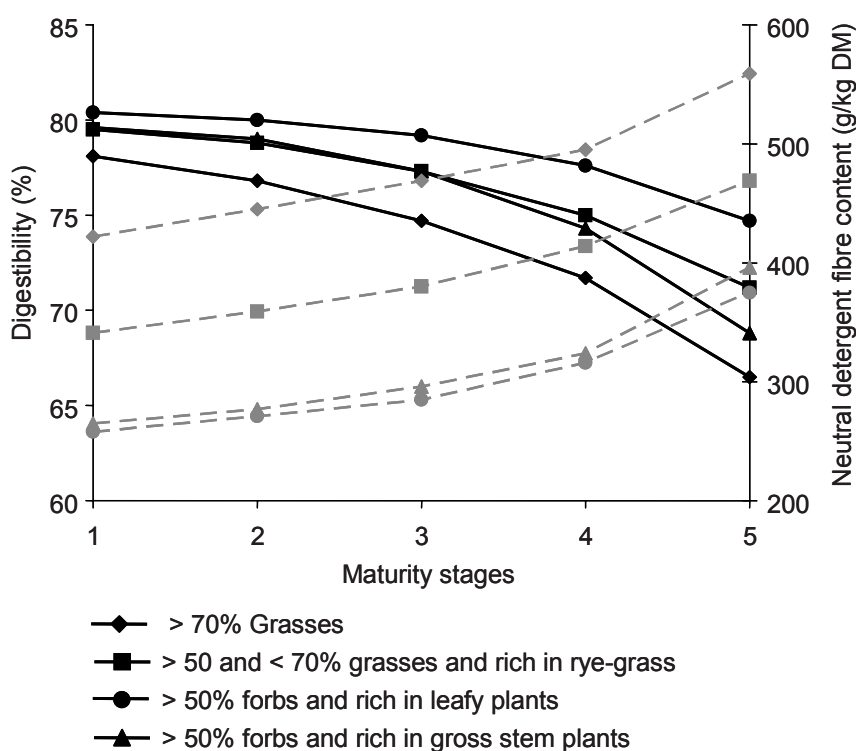


Figure 4. Evolution of digestibility (black symbols and solid lines) and cell-wall content (grey symbols and dotted lines) for different types of grasslands characterized by their botanical composition. (Adapted from Daccord *et al.*, 2006).

### How to genetically improve feed quality in complex grasslands?

Swards with complex specific composition follow the same general relationships between biomass and quality or among chemical traits as swards of a single species. Thus, it may be expected that a genetic improvement of the various components of the swards will improve the agronomic value of the complex plant communities, even though their respective abundance will be a key factor to determine the net biodiversity effect on biomass production and feed quality, and especially potential interactive effects in terms of N nutrition, mineral nutrition and animal health.



In this section, we will review some recent achievements in genetic improvements of the feed value of forage species, which may be relevant for complex swards, focussing on water soluble carbohydrates in grasses and polyphenol oxidase in perennial legumes.

#### *The water soluble carbohydrate fraction*

In perennial grasses, the water soluble carbohydrate (WSC) fraction was studied very thoroughly. On lambs (Lee *et al.*, 2001) and beef cattle (Miller *et al.*, 2001) on, high-WSC varieties of perennial ryegrass, grown as pure stands improve animal performance, thanks to a higher energy supply. It also influences the voluntary intake positively (Smit *et al.*, 2006). A higher WSC content of the diet modifies the kinetics of dry matter degradation, increases efficiency of microbial protein biosynthesis and may modify the rumen microbial population (Lee *et al.*, 2002, 2003).

When fed to animals, as silage in mixture with red clover, a high-WSC ryegrass cultivar showed a 20% increase in nitrogen utilization (Merry *et al.*, 2006).

When used in multi-species grasslands, high-WSC grass variety may have an impact on sward dynamics and on species abundance. For many grass species, a positive effect of WSC content was shown on the persistency in swards and improved cold and drought tolerance. Carbohydrate reserves are of major importance for regrowth. When comparing high- and low-WSC varieties of perennial ryegrass, Lasseur *et al.* (2007) showed that regrowth after defoliation was quicker for the high-WSC variety, which had more reserves and remobilized them to restore leaf area.

All these effects are likely to positively influence the abundance of a high-WSC variety in complex swards, mixed either with other varieties of the same species or with other species. However, at the moment, it is impossible to figure out the importance of this impact.

WSC content exhibits a large environmental effect, either due to the season or to the year, and some G x E interaction. As a consequence, this trait shows moderate broad-sense heritability, from 0.42 to 0.64 in perennial ryegrass, depending on the season and the organ (leaves or tiller bases) (Turner *et al.*, 2006). Breeding has already been successful with the release of varieties with high WSC content, such as Aberavon. To further progress, understanding the genetic basis of genetic variation is required. The first step towards this objective is the identification of the genomic zones involved in the control of this trait, i.e. the QTL (Quantitative Trait Loci) analysis.

Turner *et al.* (2006) used the WSC F2 mapping population, especially designed to investigate genetic control of fructan metabolism. QTLs were identified for WSC in leaves and tiller bases in spring and autumn, single QTLs explaining up to 38.7% of the total genotypic variation. WSC content was highly correlated with polymeric fructans. It was also pointed out that the QTLs were different for the two organs, with a main QTL for leaf WSC on linkage group 6. Significant dominance effects existed for WSC content and positive alleles were found in both parents. This means that there is a large potential for future genetic gains.

Thanks to the synteny existing among perennial grass species, recent achievements in perennial ryegrass should open new prospects to improve this trait in other species. This should be the case in cocksfoot, where the mean WSC content may be below the threshold of 7 to 16% considered adequate for silage fermentation. Investigating a set of 65 accessions of cocksfoot, Sanada *et al.* (2007) found significant differences in WSC content both in foliage and tiller bases. A more than 2-fold variation was detected for WSC content in leaves, with ecotypes from Hokkaido (Japan) exhibiting the highest content in all organs and seasons.

### *Polyphenol oxidase*

Polyphenol oxidase (PPO) is expected to reduce proteolysis and lipolysis and should thus contribute to improve feed value. This enzyme involved in the browning reaction of red clover leaves, when cut or crushed and exposed to air, where it reacts with phenol molecules. Lee *et al.* (2004) showed that this enzyme was involved in proteolysis and lipolysis processes, both being inhibited by high PPO activity. Breeding of forages with high PPO activity would be very beneficial to reduce protein and lipid losses either during ensiling (Sullivan and Hatfield, 2006) or possibly in the rumen.

Polyphenol oxidase activity was detected in 24 red clover accessions and cultivars and significant variation was detected in the range of PPO activity by Jones *et al.* (1995). The partitioning of genetic variation among and within red clover populations is not known.

For the legume species where no activity was ever detected, one possibility is to create transgenic material. This was successfully done by Sullivan *et al.* (2004) on alfalfa using three genes selected from a red clover genomic library and encoding for PPO. Even if the PPO activity of alfalfa transgenic plants was lower than in red clover, a 80% reduction in postharvest proteolysis was observed in the PPO1 transgenic alfalfa in presence of an *o*-diphenol, phenol being required as a substrate for PPO activity. This suggests that PPO activity lower than in red clover could be efficient for practical use.

Among grass species, the highest PPO activity was detected in cocksfoot and the lowest in tall fescue, 740 and 6.5 U g<sup>-1</sup> fresh weight respectively (Lee *et al.*, 2006).

If cultivars are developed with a high PPO activity, the possible consequences in complex grasslands remain unknown. Indeed, the possibility of cross reaction between PPO from one species with the protein and lipids of the whole sward is unknown. The experiments run by Lourenco *et al.* (2005) with mixtures of red and white clover and perennial ryegrass showed modification of the lipolysis activity, suggesting interaction between the various species of the swards after processing. In the analysis of a large database of rumen protein degradability measurements, the degradability of protein was found to be significantly lower for grasslands with complex species composition compared to pure grasses and pure legumes (Nozières *et al.*, 2006). Having in mind that transgenic alfalfa with a low PPO activity exhibited an 80% reduction in proteolysis, swards combining of PPO rich- and PPO-free species could be a promising issue to reduce proteolysis and a better utilization of nitrogen. Similar interaction between different components of the diets was found for the action of condensed tannins (CT). Hedqvist *et al.* (2000) showed on a set of seven varieties of *Lotus*, that despite a low mean CT content, there was a negative relationship between CT content and protein digestibility, while Burggraaf *et al.* (2004) on white clover grazed by dairy cows observed that a slight increase in CT content reduced rumen proteolysis. It was also shown that CT of *Lotus* or sainfoin have an effect on the digestibility of protein of another legume when both species were incubated together (Julier *et al.*, 2002; Aufrère *et al.*, 2005). However, when nutritional interactive effects are concerned, type of tannins, tannin content and characteristics of the other species of the diet are critical for lower protein degradability and better feed value (Robbins *et al.*, 2002).

### **Conclusions and prospects**

In this review, we have documented the variation in feed quality in swards with complex specific composition. As for monospecific swards, chemical composition and feed quality appear to be mainly influenced by the aerial biomass. It is thus influenced by the abundance of the various species. It has also been shown that genetic improvements in feed value may

easily be valorised when improved varieties are included in mixtures for sown grasslands. This is especially the case for water-soluble carbohydrates and for polyphenol oxidase. Significant progresses were also made in characterizing digestibility of grass species of semi-natural grasslands in relation with their growth strategy. The functional approach should be helpful to define grassland types and characterise their feeding value. However the role of forbs in feed quality of grasslands remains less thoroughly understood, in particular their contribution in supplying micro-nutrients of interest for animal health and quality products. This implies further investigation of animal responses to complex swards and in particular a better understanding of the interactions that can occur between plants on digestion and intake.

This opens new prospects for research and especially for modelling. Modelling would be very important to assess three issues, which are the specific abundance, the genotypic variation with each species and the consequences of management practices, and especially herbivore defoliation.

One possible outcome of modelling may be the development of decision tool kits for the managers. These would be very important in three domains that are i) choice of species and varieties for either sowing a temporary grassland or renovating a permanent sward to optimize both production and feed value, ii) assessment of the feed value of a grassland from its botanical composition, and iii) optimization of practices and especially defoliation regime to stabilize species composition and optimize production and feed value.

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## **Session 2A**

### **Food quality**

# Effects of the grazed horizon in perennial ryegrass swards on the conjugated linoleic acid concentration in milk of dairy cows

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## Abstract

Conjugated linoleic acids (CLA) are beneficial poly-unsaturated fatty acids (FA) in milk, resulting from FA in feed. Data on effects of grazing system on ingested FA are scarce, especially in relation to FA in the milk. In rotational grazing systems, amount and quality of herbage on offer differ greatly between the first and later days of a grazing cycle. The objective of this experiment was to determine FA concentrations in ungrazed and pre-grazed swards in relation to FA intake and CLA content in milk of grazing dairy cows.

Twelve cows were allocated to two treatments, i.e., starters, offered ungrazed swards and followers, offered pre-grazed grass during 8 days, and changed thereafter. Sward height, forage quality and FA in grass and milk were measured. Data were analyzed using a Latin Square design.

Leaf blade proportion of the canopy decreased gradually from ungrazed plots to 2-day grazed plots. The concentration of FA was highest ( $P < 0.001$ ) in the herbage of the starters, which produced more ( $P < 0.001$ ) CLA than the followers. A positive relation ( $P < 0.05$ ;  $R = 0.58$ ) between the CLA content of the milk and the  $\alpha$ -linolenic acid content of the grass was found. Grazing management strategies could thus modify milk FA composition.

Keywords: milk quality, fatty acid composition, herbage quality, diet selection, intake, grazing system

## Introduction

Consumers are concerned about the quality and healthiness of their food. Polyunsaturated fatty acids (FA) have beneficial health effects on cardiovascular problems. Dairy products constitute a large part of fat intake and modification of the FA profile of milk fat is desired (Jensen, 2002). Conjugated linoleic acid (CLA) is an important poly-unsaturated FA in the milk of dairy cows with beneficial health effects (Elgersma *et al.*, 2004). Forage species, cultivar, stage of growth, season and amount of fertilizer affect forage lipid composition, and can change the FA composition in the milk (Dewhurst *et al.*, 2001; Jensen, 2002; Witkowska *et al.*, 2006). However, information on grazing strategies in relation to the CLA concentration in milk is lacking. The objective of this experiment was to determine FA concentrations in ungrazed and pre-grazed swards in relation to FA intake and CLA in milk of grazing HF dairy cows.

## Materials and methods

The experimental period, 1-18 September 2005, was divided into two equal periods of eight days. The first four days were considered as an adaptation period for the animals and data from the last four days in each period were analyzed. Twelve cows were divided into two groups of 6, allocated to two treatments, i.e., starters and followers. Treatments changed in period 2. Starters were allocated daily at noon to a new ungrazed plot, while followers were allocated to the pre-grazed plot. Sward surface height (SSH) was measured each day in



ungrazed and pre-grazed plots with a falling plate meter each day at 8:30, 13:30 and 18:30 h. At the same moment, grass samples were taken with a knife, mimicking the grazed horizon of the cows. Half of this sample was oven-dried, ground to 1 mm and analyzed using near infrared spectroscopy (NIRS) for chemical composition and digestibility. The other half of the sample was frozen (-20°C) and used for FA determination (Elgersma *et al.*, 2005). Herbage dry-matter yield (DMY) and grass morphology were measured six times during the experiment in ungrazed, 24-h pre-grazed, and 48-h pre-grazed swards, by cutting three strips of 5 x 1.25 m in each plot at a fixed 4 cm cutting height. All fresh herbage was collected and weighed, samples of 250 grams were oven-dried (48 hours at 70°C) to determine DM content. DMY was related to the accompanying SSH to create a calibration line. Morning and evening milk was sampled daily, pooled, and used to determine concentrations of fat, protein and lactose. The CLA concentration of the milk fat was determined and data were analyzed using a 2 x 2 Latin Square design replicated six times, with treatment, cow and period as factors.

## Results and discussion

SSH and DMY decreased during the two days of grazing, though the standard deviation was high. The leaf blade proportion decreased gradually from ungrazed plots to 2-day grazed plots from 0.64 to 0.50 to 0.42 of DM. The crude protein concentration was higher ( $P < 0.001$ ) for starters than followers (298 vs. 255 g kg<sup>-1</sup> DM). Followers had higher concentrations of water-soluble carbohydrates ( $P < 0.05$ ), crude fibre, NDF, ADF and ash ( $P < 0.001$ ) in their diet. The concentration of total FA was higher ( $P < 0.001$ ) in the herbage of the starters than of the followers (29.5 vs. 24.2 g kg<sup>-1</sup> DM) (Table 1), mainly due to C18:3 (23.5 vs. 18.1 g kg<sup>-1</sup> DM). The proportion of C18:3 was highest ( $P < 0.001$ ) in diet of the starters (0.80 vs. 0.75), whereas proportions of C16:0, C18:0, C18:1 and C18:2 were higher ( $P < 0.01$ ) in the diet of the followers. The higher FA content in the grazed horizon of ungrazed plots confirms the hypothesis that FA are mainly located in leaf blades in upper canopy layers (Witkowska and Elgersma, unpublished results).

Table 1. Concentration of the five major fatty acids (FA; g kg<sup>-1</sup> DM) in herbage grazed by the starter and the follower group.

FA	Starter	Follower	s.e.d.	Significance
C16:0	3.06	2.81	0.06	**
C18:0	0.25	0.22	0.01	*
C18:1	0.34	0.35	0.02	ns
C18:2	2.35	2.66	0.05	***
C18:3	23.54	18.13	0.45	***
Total	29.53	24.17	0.53	***

ns: not significant, \*:  $P < 0.05$ , \*\*:  $P < 0.01$ , \*\*\*:  $P < 0.001$ .

Starters and followers did not differ in DM intake due to the large variation in estimates, mean values were 18 vs. 10 kg d<sup>-1</sup> in period 1 and 19 vs. 12 kg d<sup>-1</sup> in period 2. The average daily milk production was 20.5 kg fat-and-protein corrected milk and decreased throughout the experiment. The milk production was higher ( $P < 0.05$ ) for starters than for followers (Table 2). The milk solids did not differ between treatments. Starters had a higher ( $P < 0.001$ ) CLA concentration in milk fat as well as in total milk than followers. The starters produced almost 10 grams more ( $P < 0.001$ ) CLA per day than the followers. A positive relation ( $P < 0.05$ ;  $R = 0.58$ ) between the CLA concentration of the milk and the C18:3 concentration of the grass was found, whereas the other FA were negatively related to CLA in the milk.

Table 2. Milk production, concentrations of fat, protein and lactose in milk, concentrations of CLA in milk fat and milk, and CLA production.

	Starter	Follower	s.e.d.	Significance
Milk production (kg d <sup>-1</sup> )	21.3	19.5	1.29	*
Fat (g kg <sup>-1</sup> )	42.7	42.2	1.87	ns
Protein (g kg <sup>-1</sup> )	34.5	34.0	0.09	ns
Lactose (g kg <sup>-1</sup> )	44.2	43.6	0.05	***
CLA in fat (g 100 g <sup>-1</sup> )	2.45	1.67	0.21	***
CLA in milk (g kg <sup>-1</sup> )	1.02	0.72	0.08	***
CLA production (g d <sup>-1</sup> )	21.5	12.8	1.97	***

ns: not significant, \*:  $P < 0.05$ , \*\*\*:  $P < 0.001$ .

The outcomes of this experiment confirm that the concentration of CLA in milk responds quickly to changes in FA in the diet (Elgersma *et al.*, 2004). C18:3 in feed is a precursor for CLA in milk and this is in line with the positive relation between the C18:3 concentration in the grass on offer and the CLA concentration in milk found in this study with grazing, and earlier in an indoor feeding experiment (Elgersma *et al.*, 2003). Effects on fatty acids in milk other than CLA will be presented elsewhere.

## Conclusions

A vertical gradient in FA concentration in grass canopies was found that was related to leaf blade proportion. Cows with access to upper sward horizons had higher CLA concentrations in their milk than cows offered pre-grazed swards. Grazing strategies could thus affect milk FA composition.

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# Effects of harvest time of red clover silage on milk production and composition

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## Abstract

Twelve cows were used in a cyclic change-over experiment to examine the effects of harvest time of red clover silage on milk production and composition. Dietary treatments consisted of four red clover silages harvested from primary growths of early and late cut red clover sward and their regrowths, and of two grass silages harvested from early and late cut primary growths of timothy-meadow fescue sward. Silages were fed *ad libitum* and supplemented with 9.5 kg d<sup>-1</sup> of a concentrate. Advancing maturity of primary growth silages decreased dry matter (DM) intake of silages and energy-corrected milk yields of cows irrespective of the forage species used, the reductions being less for red clover than grass diets. Milk protein content was higher for grass than red clover while milk fat content was similar. However, compared with grass diets, red clover enhanced milk fat unsaturated fatty acid (FA) content, C18:3 n-3 in particular, and decreased that of saturated FA. Red clover diets from secondary rather than primary harvest increased silage DM intake, milk yield, milk protein and C18:3 n-3 contents. In conclusion, timing of harvest of red clover represents a potential strategy for extending harvesting date of primary growth, and modifying milk fat composition on high forage diets.

Keywords: dairy cows, red clover silage, grass silage, maturity stage, regrowth, milk fat

## Introduction

Intake and milk production potential of forages are largely dependent on plant species, maturity stage of herbage and conservation method used in silage production. During recent years competitiveness of N<sub>2</sub>-fixing forage legumes such as red clover (*Trifolium pratense*) for silage production has increased owing to potential to save N fertilization. Red clover silage fed either alone or as a part of legume-grass silage has stimulated higher DM intake and milk yield in comparison with grass silages (Bertilsson and Murphy, 2003; Dewhurst *et al.*, 2003; Vanhatalo *et al.*, 2006). Replacing grass silage with red clover silage in these studies has also often led to reductions in milk fat and/or protein concentrations, and changes in milk fatty acid (FA) composition (Dewhurst *et al.*, 2003; Vanhatalo *et al.*, 2007). However, high milk yields obtained with pure red clover silage have not consistently been attributed to a higher DM intake of red clover silage. In an earlier study (Vanhatalo *et al.*, 2005) advancing maturity decreased DM intake of grass silages but the opposite was true for pure red clover silages. While the effects of stage of maturity on DM intake and milk production of cows fed grass silage diets are fairly well documented (see e.g. Rinne, 2000) data concerning the maturity effects of red clover silage are limited. The aim of this study was to examine the effects of timing of harvest of red clover silage on silage DM intake, milk production and composition.

## Materials and methods

Experimental silages were prepared during summer 2004 from primary growths of early cut (15 June; early heading) and late cut (30 June; late heading) timothy-meadow fescue sward,

and from primary growths of early cut (1 July; early flowering) and late cut (14 July; late flowering) pure red clover sward, and their regrowths (24 August; mid-flowering or early flowering, respectively). The swards were harvested with a mower conditioner, wilted for up to 8 h (grass) or up to 48 h (red clover), and conserved with 5 l tonne<sup>-1</sup> of a formic acid based additive (760 g formic acid kg<sup>-1</sup>) in round bales wrapped with six layers of plastic.

A cyclic change-over design experiment (Davis and Hall, 1969) with four 21-day periods was conducted using twelve Finnish Ayrshire cows in early lactation. Dietary treatments consisted of the two grass silages and the four red clover silages offered *ad libitum*, each supplemented with, on average, 9.5 kg d<sup>-1</sup> concentrate mixture consisting of oats-barley (1:1) and rapeseed expellers (24%). Experimental feeds were sampled, and feed intake and milk yields of cows were recorded throughout the experiment. Milk samples were obtained from four consecutive milkings on days 18 to 20 of each period. Measurements from the last week of each period were used in statistical analyses.

## Results and discussion

Owing to unstable weather conditions during the harvesting of red clover the DM concentrations of red clover silages were much lower than those of grass silages (Table 1). All silages were well preserved with only trace amounts of butyric acid. However, red clover silages were more extensively fermented than grass silages. Consequently, comparison of silage DM intakes between the plant species may be confounded due to differences in the extent of silage fermentation.

Advancing maturity of silages decreased DM intake of silages and energy-corrected milk yields (ECM) of cows irrespective of the forage species used (Table 2), the reductions being smaller for red clover than grass silage diets ( $P < 0.01$ ). In a previous study (Vanhatalo *et al.*, 2005) advancing maturity of primary growth red clover silage was attributed to increased DM intake. However, the digestibilities of the present primary growth red clover silages were generally lower than those in the previous study. Higher ( $P < 0.001$ ) milk protein concentration for grass than red clover silage diets agreed with earlier findings (Bertilsson and Murphy, 2003; Vanhatalo *et al.*, 2005), whereas milk fat content was unchanged ( $P > 0.05$ ) in the present study.

Table 1. Chemical composition and fermentation quality of the experimental silages (n = 4).

Experimental silage	Grass		Red clover		Regrowth of red clover	
Growth stage	Early	Late	Early	Late	Early	Late
pH	4.25±0.032	4.11±0.049	4.05±0.038	4.30±0.102	4.22±0.039	4.31±0.016
Dry matter (DM), g kg <sup>-1</sup>	321±9.7	277±13.6	180±6.0	224±10.2	251±12.7	219±16.5
Crude protein, g kg <sup>-1</sup> DM	112±4.1	90±2.0	166±4.3	165±3.1	170±2.9	193±9.2
NDF, g kg <sup>-1</sup> DM	541±10.6	601±11.4	395±3.3	391±2.5	373±8.7	348±25.6
Indigestible NDF, g kg <sup>-1</sup> DM	82±1.7	162±1.1	184±7.5	203±12.4	177±4.1	141±12.2
D-value, g kg <sup>-1</sup> DM	696±2.2	619±1.7	639±2.2	592±1.0	619±1.8	651±2.5
WSC, g kg <sup>-1</sup> DM	102±14.6	45±9.1	3±2.3	12±2.3	33±9.2	8±6.0
Lactic acid, g kg <sup>-1</sup> DM	42±4.8	44±9.1	102±8.0	106±8.7	86±11.1	98±9.6
Acetic acid, g kg <sup>-1</sup> DM	9±1.0	9±1.1	27±1.8	31±3.6	20±2.0	29±2.4
Butyric acid, g kg <sup>-1</sup> DM	0.12±0.004	0.08±0.035	0.06±0.002	0.49±0.326	0.05±0.021	0.05±0.004
Ammonia N, g kg <sup>-1</sup> N	53±0.27	90±0.54	62±0.59	112±0.34	68±0.50	66±0.60

NDF = neutral detergent fibre, D-value = digestible organic matter in DM, WSC = water soluble carbohydrates.

Consistent with earlier studies (Dewhurst *et al.*, 2003; Vanhatalo *et al.*, 2007) red clover enhanced ( $P \leq 0.01$ ) milk fat monounsaturated fatty acid (MUFA) and polyunsaturated fatty acid (PUFA) contents, C18:3 n-3 in particular, and decreased ( $P < 0.05$ ) that of saturated FA as compared with grass silage diets. As found earlier (Vanhatalo *et al.*, 2007) the most pronounced increase in milk PUFA was related to the early cut red clover diet.

Further, red clover diets from secondary rather than primary harvest increased ( $P < 0.001$ ) silage DM intake, milk yield ( $P < 0.05$ ), milk protein ( $P < 0.05$ ) and 18:3 n-3 contents ( $P < 0.001$ ). This was in line with the slightly higher organic matter digestibility and lower concentration of indigestible neutral detergent fibre of the secondary than primary cut red clover silages.

Table 2. Feed intake, milk production and composition in cows fed grass- and red clover - silage diets.

Experimental silage	Grass		Red clover		Regrowth of red clover		SEM	Statistical
Growth stage	Early	Late	Early	Late	Early	Late		significance
Intake								
Silage dry matter, kg d <sup>-1</sup>	14.3	8.3	10.7	9.0	12.8	11.8	0.67	***
Total dry matter, kg d <sup>-1</sup>	23.0	17.0	19.1	17.4	21.3	20.2	0.67	***
Production								
Milk, kg d <sup>-1</sup>	28.6	25.5	28.3	28.4	29.1	29.7	1.05	***
ECM, kg d <sup>-1</sup>	29.9	26.6	29.2	28.6	29.7	30.1	1.11	**
Fat, g kg <sup>-1</sup>	43.9	44.3	44.2	41.2	42.2	41.6	1.56	
Protein, g kg <sup>-1</sup>	35.1	34.9	33.6	33.9	34.8	34.0	0.62	**
Lactose, g kg <sup>-1</sup>	45.4	44.9	45.0	45.9	45.7	45.7	0.43	***
Urea, mg (100 ml) <sup>-1</sup>	25	31	38	37	35	42	2.1	***
Milk FA, g (100 g FA) <sup>-1</sup>								
C18:2 (n-6)	1.89	2.08	2.36	2.23	2.43	2.34	0.075	***
C18:3 (n-3)	0.44	0.48	0.87	0.67	0.92	0.93	0.048	***
Saturated FA	71.3	68.0	67.2	67.3	68.5	67.8	0.63	***
MUFA	23.8	27.0	27.2	27.3	25.5	26.1	0.59	***
PUFA	2.78	3.09	3.83	3.48	3.89	3.84	0.113	***

ECM = energy corrected milk yield, FA = fatty acids, MUFA = monounsaturated FA, PUFA = polyunsaturated FA. Statistical significance of the experimental treatments: \*\*\*  $P < 0.001$ , \*\*  $P < 0.01$ . SEM = standard error of the mean.

## Conclusions

Timing of harvest of red clover represents a potential strategy for extending harvesting date of primary growth forages without major compromise in milk production, and modifying milk fat composition when high forage diets are used in dairy cow feeding.

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# Influence of botanically different forages on rumen, milk and tissue fatty acid metabolism

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## Abstract

Three *in-vivo* and one *in-vitro* experiments were performed to assess the effect of feeding botanically diverse (BD) forages on the fatty acid (FA) composition of dairy milk and lamb intramuscular (IM) fat, and the potential 'origin' of differences in the latter based on precursor supply and changes in the endogenous ruminant FA metabolism. A general increase in milk and meat polyunsaturated FA (PUFA) and conjugated linoleic acid (CLA) levels was observed upon feeding BD forages. These observations were associated with higher rumen C18:1 t11 proportions in BD forage fed animals. This could not always be explained by differences in the dietary precursor supply of PUFA. Plant secondary metabolites present in specific herbs of botanically diverse forages are possible candidates to modify the hydrogenating rumen microbial population, although the current *in-vitro* screening did not provide any evidence for saponins, quercetin or eugenol, plant secondary metabolites present in *Fabaceae*, *Asteraceae* and *Ranunculaceae* families, respectively, to modify the rumen FA metabolism.

Keywords: biohydrogenation, botanical composition, fatty acid metabolism, milk, meat

## Introduction

Diets rich in leguminous plant species, in particular white and red clover, have been shown to improve the ruminant milk and meat FA profile in terms of human nutritional recommendations (Dewhurst *et al.*, 2006). Alpine/mountain systems, characterized by high forage proportions and botanical diversity, have also been described to increase the n-3 PUFA and CLA c9t11 (although less consistent) contents in ruminant fat (Lourenço *et al.*, 2008). As no specific mechanisms have yet been suggested to explain these differences, the *in-vivo* studies aimed at assessing the effect of feeding botanically diverse forages (fresh or conserved) on the fatty acid composition of ruminant milk or meat and to relate it to the rumen FA profile. Additionally, an *in-vitro* test was performed in an attempt to identify secondary plant metabolites which might modify rumen FA metabolism.

## Materials and methods

In the first experiment, 4 lactating ( $249 \pm 79$  days in lactation) and rumen cannulated Holstein cows were used in a 4 x 4 Latin square design (Lourenço *et al.*, 2005) from which 2 dietary treatments are reported: 100% intensively managed ryegrass silage (100 IMS) vs. 40% IMS and 60% semi-natural biodiverse silage (60 BS – 34% herbs; 11% legumes and 55% grasses). Both groups received standard dairy concentrate (4.5 kg DM d<sup>-1</sup>). Samples of the feed, rumen and milk were collected and analysed for their FA profile. In the second experiment, 2 dietary groups (7 male lambs ( $22.3 \pm 1.2$  kg) per group) are reported: botanically diverse (BD – 21% herbs; 14% legumes and 65% grasses) vs. intensive ryegrass (IR). Lambs were grazing exclusively on these pastures for 84 days before slaughter. Pasture, rumen contents and *Longissimus* muscle samples were collected (Lourenço *et al.*, 2007b). In the third experiment (Lourenço *et al.*, 2007a), 2 dietary groups (6 male lambs ( $29.6 \pm 1.5$  kg) per group) are

compared: intensive ryegrass silage (IRS) vs. botanically diverse silage (BDS – similar botanical composition as BD pasture in experiment 2). Lambs were fed for 76 days before slaughter and sampling. Both groups received an additional amount of wheat and barley, balanced according to their net energy and digestible protein contents. A final experiment aimed at screening, with continuous culture fermenters, the effect of three different plant secondary metabolites (eugenol (EUG, 250 mg L<sup>-1</sup>), triterpene saponin (1000 and 500 mg L<sup>-1</sup>) and quercetin (500 and 250 mg L<sup>-1</sup>)) on rumen PUFA biohydrogenation. Doses tested were based on literature data and for triterpene saponin and quercetin two doses were tested. A negative control (CON) was included. For more details on the experimental design, analytical methodology and statistical analysis from each experiment we refer to the individual papers (Lourenço *et al.*, 2005; 2007a,b).

## Results and discussion

In the first experiment, milk CLA c9t11 proportions of cows fed diet 60 BS were almost doubled compared with milk of cows fed 100 IMS, despite the reduced dietary supply of C18:3 n-3 from 60 BS (Table 1). The higher milk CLA c9t11 proportions were associated with a partial inhibition of the rumen biohydrogenation of dietary PUFA, as suggested from the accumulation of C18:1 t11 and the reduced C18:0 proportions in the rumen pool samples (Table 1). Differences in milk C18:3 n-3 were small, reflecting higher recoveries of dietary C18:3 n-3 when feeding 60 BS (5.11 vs. 7.31 g milk C18:3 n-3/100 g feed C18:3 n-3 for diets 100 IMS and 60 BS, respectively).

Table 1. Forage, rumen, milk and IM proportions of individual FA (g/100 g FA methylesters (FAME)) of Experiments 1, 2 and 3.

		Experiment 1			Experiment 2			Experiment 3		
		100 IMS	60 BS	SEM	IR	BD	SEM	IRS	BDS	SEM
Forage intake (kg DM d <sup>-1</sup> )		13.9	13.0	1.20	<i>ad libitum</i>		-	1.16	1.02	0.075
Forage	C18:2 n-6	11.0	14.6*	0.579	13.9	18.2*	0.778	18.6	28.1*	0.776
	C18:3 n-3	48.0	38.6*	0.874	57.2	51.7*	1.67	49.7	33.2*	1.28
Rumen	C18:1 t11	2.14	3.71*	0.093	4.47	7.29*	0.365	1.05	2.51*	0.278
	C18:0	45.2	39.4*	0.542	57.6	53.1*	0.787	53.8	40.3*	2.17
Milk or IM fat	C18:1 t11 <sup>†</sup>	0.469	0.822*	0.017	2.74	2.22	0.258	0.353	0.567	0.095
	CLA c9t11	0.259	0.438*	0.012	0.903	0.897	0.097	0.301	0.595*	0.078
	C18:3 n-3	0.612	0.590	0.013	2.59	2.64	0.235	1.62	1.53	0.151
Indices for elongation and desaturation	C20:5 n-3/C18:3 n-3	-	-	-	0.273	1.02*	0.103	0.489	0.702(*)	0.083
	C22:5 n-3/C18:3 n-3	-	-	-	0.271	0.993*	0.094	0.570	0.868*	0.102
	C22:6 n-3/C18:3 n-3	-	-	-	0.131	0.161	0.012	0.166	0.178	0.017
Milk production (kg d <sup>-1</sup> )		21.9	20.5	0.917	-	-	-	-	-	-
Average daily live weight gain (g d <sup>-1</sup> )		-	-	-	165	36.4*	12.2	154	54.5*	17.5

\*, (\*) Differences (\*  $P < 0.05$ ; (\*)  $P < 0.1$ ) reported between botanically diverse and ryegrass forages within each of the 3 different experiments; <sup>†</sup> For Experiment 2, these proportions represent the sum of all C18:1 *trans* isomers identified in the IM fat

In the second and third experiments, higher C18:1 t11 proportions in the rumen contents of BD grazing and BDS animals were observed (Table 1), again suggesting a partial inhibition of the rumen biohydrogenation. In both experiments, higher indices for elongation and desaturation activity in the IM fat of BD grazing animals and BDS animals were observed (Table 1), suggesting some stimulation of elongation and desaturation to long chain FA (Lourenço *et al.*, 2007a,b). The 'origin' of the general increase in PUFA and CLA contents in milk and meat through BD forages could not always be explained by differences in the dietary precursor supply of PUFA. Potentially, plant secondary metabolites present in specific herbs of botanically diverse forages might play a role in the modification of the rumen PUFA



metabolism. Indeed, some plant secondary metabolites show rumen methane inhibitory properties which is of particular interest, as a concomitant inhibitory effect on rumen methanogenesis and PUFA biohydrogenation has been observed earlier for synthetic additives, such as monensin or dietary supplements, such as fish oil or microalgae (Lourenço *et al.*, 2008). As no response studies describing the effect of plant secondary metabolites on rumen PUFA biohydrogenation are available, an *in-vitro* screening was performed.

Table 2. Proportions of C18:1 t11 (g/100 g of FAME) in the effluent of continuous culture fermenters and C18:3 n-3 apparent biohydrogenation (g C18:3 n-3 hydrogenated/100 g C18:3 n-3 input) when supplying to the rumen simulator the different plant metabolites.

	CON	Saponin		Quercetin		EUG	SEM
Dose	-	1000 mg/ L	500 mg/L	500 mg/L	250 mg/L	250 mg/L	
C18:1 t11	13.7	13.3	13.5	14.8	13.7	14.0	1.50
<i>Apparent biohydrogenation</i>							
C18:3 n-3	85.1	84.8	86.2	85.8	84.9	83.8	0.027

At the doses tested in this study, none of the plant secondary metabolites modified microbial activity or caused shifts in the extent of rumen biohydrogenation (Table 2).

## Conclusions

BD forages have the potential to increase the PUFA content of ruminant milk and meat. This effect was associated with changes in the rumen and endogenous animal FA metabolism. Plant secondary metabolites might be the 'origin' for the observed differences, but the current *in-vitro* screening did not provide any evidence for this.

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# Growth and carcass quality of suckler calves grazing at the farm or in the mountains

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## Abstract

The objective was to examine the effect of mountain grazing on the growth and meat quality of suckler calves. Suckler calves with their dams were raised on either cultivated pasture (C) or free range on mountain/forest pasture (M) on 4 and 5 commercial farms in 2006 and 2007, respectively. The M included different vegetation types from open, grass-dominated meadows to forests with patches of grasses, herbs and shrubs. All calves were slaughtered at the end of the grazing season at an average age of 203 (2006) and 193 days (2007). On average, there was no effect of pasture type on production traits except for carcass fatness, which was higher on M than on C. In 2006, significantly higher live weight gain and carcass weights were found in the C treatment, whilst in 2007 the reverse effect was found. From first-year results, total lipid content in tissue of *M. Longissimus dorsi* was higher in the M treatment (12 vs. 14 g kg<sup>-1</sup>,  $P < 0.05$ ). There were no other differences in meat quality between treatments.

Keywords: beef cattle, calf, carcass quality, pasture, production system

## Introduction

Historically, cattle were the most important domestic animal species, in terms of feed intake and production, used for forest and mountain grazing in Norway. It was as late as the middle of the last century that sheep became more important. Locally, forest and mountain grazing with cattle is still important, and interest is increasing with more specialized beef production and niche marketing. Forest and mountain grazing may be attractive economically as it is stimulated by the authorities as a measure to maintain a desired cultural landscape and due to low feed costs. More recently it has been asked to what extent mountain and forest grazing affects the nutritional quality of the meat, as effects of pasture type on dairy milk quality and meat quality in cattle and sheep have been demonstrated (Larick *et al.*, 1987; Adnøy *et al.*, 2005; Leiber *et al.*, 2005). The objective of this study was to investigate the effect of forest and mountain grazing on growth and carcass and meat quality of suckler calves.

## Materials and methods

On each of nine commercial farms, four in 2006 and five in 2007, 10-12 suckling calves with their dams were raised on either cultivated pasture (C) or on mountain/forest pasture (M) in the municipalities Gausdal, Lillehammer and Øyer, south-east Norway. Two farms participated both years. Within farm the experimental animals were divided randomly into the two experimental groups taking birth date, birth weight, parity of the dam and sex into account. All calves were weighed at birth, the day of turn-out to M, and the day at gathering from M. The calves were born at end of February / early March, and average age at slaughter was 203 (SD 31) and 193 (SD 25) days in 2006 and 2007, respectively. In M the animals were grazing

free-range in vegetation types that varied from open, grass-dominated meadows to forests with patches of grasses, herbs and shrubs between the trees (mainly *Betula* spp. and *Picea abies* L.), at altitude range 600–1200 m a.s.l. (tree limit is at ca. 1,050 m). The C was fertilized, continuously grazed permanent pastures located close to the farm. Important species were *Poa pratense* L., *Agrostis capillaris* L., *Deschampsia cespitosa* L., *Achillea millefolium* L., *Rumex acetosa* L., *Ranunculus acris* L. However, on all farms in 2006 and on two farms in 2007, calves in C were grazing on *Lolium multiflorum* Lam. (5 farms) or *Dactylis glomerata* L. (1 farm in 2006) dominated pastures for at least 3 weeks before slaughter. On all farms, except one with Aberdeen Angus (both years) and one with Charolais (1 year), the offsprings were crosses of large breeds (Simmental, Charolais and Norwegian Red). All calves within farm were slaughtered at the same date within 1 week after gathering from M. Samples of the *M. longissimus dorsi* and of subcutaneous fat were collected within 24 hours after slaughter and analysed chemically and meat colour measured instrumentally. Data from each year were analysed using a mixed model where pasture type, farm, sex, age at turn-out to M (covariate) and live weight gain from birth to turn-out to M (covariate) were included as fixed effects and animal within herd as a random effect. In addition, the data from the two farms that participated both years were subjected to a separate analysis where the effect of year also was included in the model. Data are presented as LS means.

## Results and discussion

Live weight gain during the grazing period, live weight at slaughter and carcass weight were significantly higher on C than on M in 2006 while the opposite results were obtained in 2007 (Table 1). Results from the two farms participating both years revealed significant year by pasture type interaction for most production traits; the production performance was higher on C than on M in 2006, and vice versa in 2007 (Table 2). The only exception was carcass fatness that was consistently higher on M than on C (Table 2). Higher carcass fatness was obtained without any difference in growth rate or carcass weight.

Table 1. Effect of pasture type on live weight at slaughter, carcass weight and daily live weight gain (LWG) during the grazing period and carcass traits of suckler calves grazing on cultivated pastures (C) or in forest/mountain (M) in two grazing seasons (2006 and 2007).

	2006				2007			
	Pasture type		SEM <sup>a</sup>	Signif. <sup>b</sup>	Pasture type		SEM <sup>a</sup>	Signif. <sup>b</sup>
	C	M			C	M		
N	19	23			30	30		
Live weight, kg	246	230	3.3	**	214	225	2.7	**
LWG, g day <sup>-1</sup>	1073	948	29.4	**	984	1104	26.5	**
Carcass weight, kg	128	123	1.8	(*)	112	117	1.8	*
Conformation <sup>c</sup>	6.5	6.1	0.16	ns	5.7	6.0	0.21	ns
Fatness <sup>d</sup>	4.6	5.1	0.30	ns	3.5	4.0	0.16	*

<sup>a</sup>SEM = standard error of the mean.

<sup>b</sup>Statistical significance; (\*),  $P < 0.1$ ; \*,  $P < 0.05$ ; \*\*,  $P < 0.01$ , ns, not significant

<sup>c</sup>EUROP system: P- = 1, P = 2, P+ = 3, O- = 4, O = 5, O+ = 6, R- = 7, R = 8, R+ = 9

<sup>d</sup>EUROP system: 1- = 1, 1 = 2, 1+ = 3, 2- = 4, 2 = 5, 2+ = 6, 3- = 7 ... 5+ = 15

The first year results are in accordance with Gravir (1962) who found that cattle grazing on lowland pastures had greater live weight gains than those grazing in the mountains. It is likely that the contrasting results in the two years were due to differences in forage allowance and quality on the M pasture between years. The weather conditions were very different; precipitation was about 30% higher and mean daily temperature 2.2°C lower in the 3-month M grazing period in 2007 compared with 2006. Cool summers with high rainfall delay plant maturity, prolong access to forage with high quality and improve plant production, and it has been found that the carcass weight of moose that graze in forest in regions with dry climate is

higher after a summer with high precipitation and low temperature than vice versa (Saether, 1985). During periods of poor pasture growth, animals on C were supplemented with silage and the yearly variation is evened out. In addition, animals on M had higher energy requirements for movement as they had to walk further for foraging.

Table 2. Effect of year (2006 or 2007) and pasture type on live weight, carcass weight, daily liveweight gain (LWG) during the grazing period and carcass traits of suckler calves grazing on cultivated pastures (C) or in forest/mountain (M).

	2006		2007		SEM <sup>a</sup>	Signif. <sup>b</sup>		
	C	M	C	M		Year	Pasture	Year × Pasture
N	11	12	12	12				
Live weight, kg	242	227	233	244	4.4	ns	ns	**
LWG, g day <sup>-1</sup>	970	830	968	1097	42.5	**	ns	**
Carcass weight, kg	128	123	120	127	2.8	ns	ns	*
Conformation <sup>c</sup>	6.3	5.9	5.5	6.4	0.29	ns	ns	(*)
Fatness <sup>d</sup>	4.2	4.5	5.3	6.7	0.37	ns	*	ns

<sup>a, b, c, d</sup> See Table 1 for explanation.

Males had significantly higher growth rate, live weight at slaughter and slaughter weight in the first year, and lower fatness in both years than the female calves (data not shown). Farm had significant effect on all production traits, which is, to a large extent, a cattle-breed effect (data not shown). However, other management effects cannot be excluded.

Chemical analysis of the meat and subcutaneous fat is available, at present, only from the first year. Muscle-fat content was significantly higher ( $P < 0.05$ ) in M (14 g kg<sup>-1</sup>) than in C (12 g kg<sup>-1</sup>), and proportion of polyunsaturated fatty acids (PUFA) of total FA in the subcutaneous fat was higher ( $P < 0.001$ ) on C (47 mg 100 g<sup>-1</sup>) than M (41 mg 100 g<sup>-1</sup>). Otherwise, there were only minor differences in meat fatty acid composition (data not shown). Average level of PUFA was high (138 mg 100 g<sup>-1</sup>), the n-6/n-3 FA ratio low (1.7) and PUFA/saturated FA ratio high (0.32) compared with previous studies, but in accord with beef production systems based on pasture and in young and lean animals with low fat content (Scollan *et al.*, 2006).

## Conclusions

Compared with grazing cultivated pastures, suckler calves grazing free range in mountain or forest with their dams may have similar growth rates during the grazing period and similar carcass weights and conformation but higher fatness. First year results show no differences in meat quality in terms of fatty acid composition.

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# Volatile compounds of Alpine vegetation as markers for the traceability of ‘grass-deriving’ dairy products

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## Abstract

Volatile compounds, particularly terpenes, are potential markers for traceability of dairy products. Results are presented on the identification of compounds transferred from alpine clover (*Trifolium alpinum*) vegetation type to dairy products. Representative samples of the herbage mass exploited by dairy cows as well as of some single representative species were harvested at two sites in the south-western Alps (Piedmont, Italy). Essential oils extracted by steam distillation of pure species and bulk herbage samples were analysed by GC/FID and GC/MS. In addition, milk samples from dairy cows exploiting the vegetation type were collected and analysed using the same technique used for vegetation. Results showed that the volatile profile of the vegetation type can be retraced to that of the individual studied species, and that milk may contain terpenes derived from grazed species.

Keywords: dairy products, GC/MS, grazing, traceability, volatile profile

## Introduction

Diversification based on scientific evidence between niche mountain and mass industrial products is an important way to promote pastoral exploitation of marginal grazing lands and to overcome the structural difficulties of mountain farming systems. With this view, the characterization of pastoral vegetation in terms of volatile organic compounds (VOCs) and the individualization of ‘plant biomarkers’ are the basis for the traceability and for the subsequent valorisation of ‘grass-fed’ dairy products. This paper reports preliminary results about the identification of compounds transferred from alpine clover (*Trifolium alpinum*) vegetation types to the milk used to produce alpine cheese. Such types are of great interest for cheese production and their VOCs profile has not been investigated up to now.

## Materials and methods

In the Piedmont Alps two homogeneous swards with an abundance of alpine clover were selected. Vegetation was surveyed with point quadrat methods and classified according to Cavallero *et al.* (2007) highland pasture categorization. Swards were located at Alpe Gianna (G) and Alpe Valcavera (V), at an altitude ranging between 2,250 and 2,350 m asl., and an aspect between NW and E. They were exploited by dairy cows during one 8-day grazing period in 2005-6 at site G, and during two 8-day grazing periods in 2006 at site V. Samples representative of the complexity of vegetation, as well as samples of the principal species (*T. alpinum*) and species rich in secondary metabolites (*Alchemilla alpina*, *Potentilla grandiflora*, at site G) were collected. Plants were selected inside homogeneous areas of about 100 m<sup>2</sup>, just before exploitations carried out, at seed ripening of the dominant species (BBCH stage 75-79; Bleiholder *et al.*, 1997). To reduce soil contamination the height of cut was set at 1 cm above the soil surface, which also corresponds to the minimum height at which plants are grazed by animals. Samples (3 replications x 150 g at each site x grazing period) were put into a glass vessel and immediately refrigerated. A sub-sample of 80 g was then charged with

preservatives (100 ml of dichlormethan) and stored at 4°C until analysis. Milk samples were collected after 5 days during which dairy cows exploited solely the two swards in order to allow rumen adaptation (Viallon *et al.*, 1999). After morning milking, 200 ml of bulk milk were put into a glass vessel and immediately refrigerated. Samples were charged with preservatives (100 ml of diethyl ether) and stored at -18°C until analysis. Vegetation and milk samples were both steam distilled for the isolation of essential oils, which were subsequently analysed with gas chromatography / flame ion detection – mass spectrometry (GC/FID-MS). Essential oil components were identified by retention indices (RI), mass spectra, and literature data. Details on analytical procedures are reported in Lombardi *et al.* (2007).

## Results and discussion

Alpine clover swards at sites G and V belonged to *T. alpinum* and *Carex sempervirens* type (Cavallero *et al.*, 2007). Alpine clover was always the most abundant species (Table 1), generally followed by oligotrophic species, such as *Nardus stricta*, *C. sempervirens*, *Avenella flexuosa* and *A. alpina*. The percentage specific contribution of *Apiaceae*, *Compositae*, *Plantaginaceae* and *Rosaceae* families, whose species are recognized as biomarker sources in the literature (Mariaca, 1997), was 16% at site G, and less than 2% at site V. Specific contribution of *Poaceae*, less rich in secondary metabolites, was 24% and 40%, at sites G and V respectively. Consequently, the potential for plant biomarkers should have been higher at site G than site V.

Table 1. Percent specific contribution (CS %) to vegetation composition at sites G and V.

Species	G		V		Species	G		V	
	CS %	SD	CS %	SD		CS %	SD	CS %	SD
<i>Poaceae</i>					<i>Fabaceae</i> and other families				
<i>Agrostis alpina</i>			1.3		<i>Trifolium alpinum</i>	46.9	11.5	36.6	5.6
<i>Agrostis tenuis</i>	7.8	7.4			<i>Alchemilla alpine</i>	17.9	7.1		
<i>Anthoxanthum alpinum</i>	5.2	3.9	0.3	0.5	<i>Antennaria dioica</i>			1.7	3.0
<i>Avenella flexuosa</i>	3.0	0.5	9.3	2.0	<i>Carex sempervirens</i>	5.3	3.2	11.9	8.3
<i>Festuca nigrescens</i>	6.8	3.7	3.0	2.7	<i>Hieracium x auriculiforme</i>			1.2	2.1
<i>Festuca gr. ovina</i>			1.9	1.4	<i>Luzula gr. campestris</i>	1.1	1.9		
<i>Nardus stricta</i>	3.5	2.5	22.1	3.6	<i>Polygonum bistorta</i>	1.1	1.0		
<i>Poa alpina</i>			2.2	0.4	<i>Potentilla crantzii</i>			1.4	1.0
					<i>Potentilla grandiflora</i>	1.7	2.0		
Total <i>Poaceae</i>	26.3	-	40.1	-	Total other species	74.0	-	52.8	-

The analysis of chromatograms allowed the identification of about 150 compounds belonging to 11 chemical classes. According to the literature, among the volatile compounds that occur in vegetation, only terpenes, phenolics and miscellaneous, transit through herbivore rumens unaltered and are helpful for traceability (Mariaca *et al.*, 1997; Viallon *et al.*, 1999; Prache *et al.*, 2005). With regard to such chemical classes, 28 terpenes and several other phenolics and miscellaneous compounds were individualized at site G in bulk herbage samples, pure species and milk (Table 2). At site V, a lower number of terpenes (17) and other classes of compounds was detected. Bulk samples from site G were richer in potential food chain tracers when compared with the ones from site V (35 vs. 17). Such a difference might be related to a greater presence of species belonging to secondary metabolites-rich families. Also the number of compounds detected in *T. alpinum* as pure species was higher at site G (26) than at site V (16). Consequently, at both sites VOCs composition was noticeably influenced by the alpine clover volatile profile: only 5 compounds at site G and 1 at site V were detected in bulk samples, and not in the alpine clover pure species. Among such compounds, only two were supplied, one each by *A. alpina* and *P. grandiflora*, species of the *Rosaceae* family. One of

the two compounds (myrtenol) was probably present also in other species, but was not detected because of a superimposition between its GC and  $\alpha$ -terpineol peaks.

Concerning milk, 12 terpenes were detected for site G, while only 6 for site V. None of them was identified at both sites but  $\beta$ -caryophyllene and phytol, which are widespread in vegetation. Neither phenolics, nor miscellaneous compounds, were identified in the milk. Nevertheless, the hypothesis that some terpenes and other potential tracers were not identified because of a very low concentration in milk, resulting in undetectable peaks in chromatograms, could not be rejected.

Table 2. Number of potential tracers identified in *Trifolium alpinum* bulk herbage samples, pure species and milk at sites G and V (Ta: *T. alpinum*; Aa: *A. alpina*; Pg: *P. grandiflora*).

site	G	G	G	G	G	V	V	V
compounds	bulk	Ta	Aa	Pg	milk	bulk	Ta	milk
monoterpenes	15	9	7	6	4	7	7	3
sesquiterpenes	8	6	1	1	7	4	3	2
other terpenoids	2	2	1	1	1	2	2	1
phenolics	5	5	1	2		2	2	
miscellaneous	5	4	1			2	2	
total	35	26	11	10	12	17	16	6

## Conclusions

Bulk herbage samples from alpine clover swards have been analysed for the first time using a methodology (mowing + stem distillation + GC-FID/GC-MS analyses) useful to simulate exploitation by grazing herbivores and manage a representative herbage mass.

Vegetation VOCs profile is rather complex: a remarkable number of compounds relevant to traceability of dairy products was detected. The comparison between VOCs profile of bulk samples and pure species shows that the VOCs profile of vegetation is affected by the dominant species (*T. alpinum*), even if *Fabaceae* species are generally considered poor in volatiles (Mariaca *et al.*, 1997). Several compounds transfer from herbage to milk, with differences between sites. Whether there is a site effect on compound transfer, or an effect of analytical methods on compounds detection, is still uncertain. The probability of peak overlapping in chromatograms is high because of the presence of a large number of compounds, and chromatogram readability may be compromised.

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# Organic selenium providing by selenized fertilization in grazed grass, grass and maize silage for beef and dairy cows

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## Abstract

Selenium (Se) is not essential for grass growth but Se content in forage is needed to cover the cattle requirements. In Belgium, the Se content in locally produced feedstuffs is low, resulting in Se deficiencies and thus metabolic disorders in cattle herds. The aim of these studies was to increase the Se contents in feedstuffs with Se-enriched fertilizers, and thus, to improve Se status in animals and Se content in milk. The fields and pastures used in a trial with Belgian Blue cattle were split into two groups (control and Se). The use of Se-enriched fertilizer increased the Se content in grass of grazed pastures by 4.9 times (246.6 vs. 50.6  $\mu\text{g kg DM}^{-1}$ ), in grass silage by 3.2 times (196.2 vs. 61.8  $\mu\text{g kg DM}^{-1}$ ) and in maize silage by 3.2 times (70.0 vs. 21.8  $\mu\text{g kg DM}^{-1}$ ). In the beef trial, the blood Se content measured by the activity of glutathione peroxidase in red blood cells remained always low (27  $\mu\text{g l}^{-1}$ ) in the control cows, while it increased in the Se group to reach 70-90  $\mu\text{g l}^{-1}$  (normal values). In the dairy trial, the Se-enriched fertilization allowed a quadrupling of the Se content in milk of grazing dairy cows (6.5 vs. 24.8  $\mu\text{g l}^{-1}$ ).

Keywords: selenium, organic, fertilizer, milk, grass, maize

## Introduction

Selenium (Se) is not essential for grass growth but Se content in forage is needed to cover the cattle requirements. It is a trace element of importance in animals owing to its implication in many biological processes such as antioxidant mechanisms, immune response, reproduction system, thyroid metabolism and anti-cancer processes. Organic forms of Se are more efficiently absorbed than inorganic forms (Fairweather-Tait, 1997).

In plants, soil selenate is taken up, reduced to selenite and forms Se-amino acids analogous to S-amino acids which are often incorporated into proteins (Mikkelsen *et al.*, 1989). However, in Belgium, soil Se is unavailable for plants so that the Se content in locally produced feedstuffs is low with, as a result, Se deficiencies and thus metabolic disorders in cattle herds. The aim of these studies was to increase the Se contents in feedstuffs with Se-enriched fertilizers, as selenate, and thus, to improve Se status in animals and Se content in milk.

## Materials and methods

Since 2002, fertilizers enriched with Se as selenate were applied on pastures and arable lands of the Experimental Station of the Veterinary Faculty of Liege University (Se group). In the pastures for grass silage production (harvesting in May and in July each year), 3 g Se ha<sup>-1</sup> were applied at the beginning of the season and after each cut. In grazed pastures, 3 g Se ha<sup>-1</sup> were applied at the beginning of the season and every 5 or 6 weeks; there were 4 or 5 applications each year. In maize, there was only one application of 8 g ha<sup>-1</sup> at sowing. The management was similar in the control plot but there is no Se addition (control group). The Belgian Blue suckler herd (35 cows at the beginning) was divided in two groups, the Se and the control groups. The animals were always affected to the same group during the whole trial. During the grazing season, the animals did not receive any supplement and grazed in a

continuous set stocking system from May until October. During the winter period, they were offered a diet composed of 30% dry matter (DM) as grass silage, 30% DM as maize silage and 14% DM as barley, these three feedstuffs being produced with or without Se-enriched fertilizer, except in years 1 and 2 for the maize which was fertilized without Se. Dried lucerne and sugar beet pulp were also added to the winter feed. The trial started in the winter period (year 1).

Se contents were determined in grazed grass (first 3 years), grass silage (4 years), maize (years 3 and 4) and barley. The Se status of the animals was expressed as whole blood Se content measured by the activity of glutathione peroxidase in red blood cells on blood samples obtained every 2 months. It is usually used as a good Se status measurement in cattle. The dairy cow trial was carried on in pastures during a short period. Pastures were grazed in a rotational system with 5 paddocks by 45 dairy cows. One of the paddocks was fertilized with Se (3 g ha<sup>-1</sup>) and the others without Se. The cows stayed 12 days in June in the Se paddock. Se contents were determined in tank milk before the entry into the Se paddock, during the stay in the paddock and one month after the removal from the Se paddock.

## Results and discussion

The results about the Se contents relative to the first 4 years in grass and maize silages and grazed grass are given in Table 1.

Table 1. Concentration of Se ( $\mu\text{g kg DM}^{-1}$ ) in different feedstuffs grown with or without Se enriched fertilizer.

	Control			Selenium		
	Grazed Grass	Grass silage	Maize silage	Grazed grass	Grass silage	Maize silage
Year 1	53.8 $\pm$ 14.1	54.2 $\pm$ 20.8		265.6 $\pm$ 113.4	165.1 $\pm$ 37.6	
Year 2	49.6 $\pm$ 18.8	50.11 $\pm$ 11.0	-	230.6 $\pm$ 120.5	183.6 $\pm$ 54.2	-
Year 3	48.4 $\pm$ 16.3	56.0 $\pm$ 17.9	27.3 $\pm$ 1.5	243.7 $\pm$ 110.1	211.8 $\pm$ 83.3	102.3 $\pm$ 7.6
Year 4	-	86.9 $\pm$ 9.0	16.4 $\pm$ 3.6	-	224.1 $\pm$ 36.0	37.8 $\pm$ 2.7

The use of Se-enriched fertilizer increased the Se content in grass of grazed pastures by 4.9 times (246.6 vs. 50.6  $\mu\text{g kg DM}^{-1}$ ), in grass silage by 3.2 times (196.2 vs. 61.8  $\mu\text{g kg DM}^{-1}$ ) and in maize silage by 3.2 times (70.0 vs. 21.8  $\mu\text{g kg DM}^{-1}$ ). The blood Se content of cows remained low during the whole trial (27  $\mu\text{g l}^{-1}$ ; Figure 1) in the control cows, while it increased in the Se group to reach a range between 50 and 90  $\mu\text{g l}^{-1}$ ; 70  $\mu\text{g l}^{-1}$  being considered as a normal value.

In the dairy trial, the Se-enriched fertilization allowed the quadrupling of the Se content in milk of grazing dairy cows (Figure 2). Initial milk Se content was 6.5  $\mu\text{g l}^{-1}$ . The mean milk Se content when the cows were in the Se paddock was 24.8  $\mu\text{g l}^{-1}$ . The higher value was reached 5 days after the entry in the Se paddock at 28.7  $\mu\text{g l}^{-1}$ . Se milk content decreased to 20, 12 and 11  $\mu\text{g l}^{-1}$  respectively 2, 4 and 6 days after the removal of the Se paddock. After one month, Se content came back at the initial level. The values in the Se paddock are higher than those observed by Pehrson *et al.* (1999) with suckler cows receiving Se yeast products (17.3  $\mu\text{g l}^{-1}$ ).

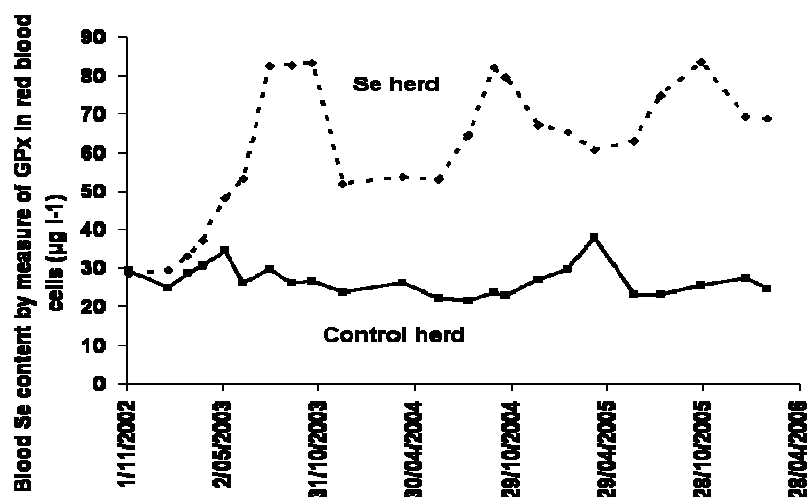


Figure 1. Evolution of the whole blood Se concentration ( $\mu\text{g l}^{-1}$ ) measured by glutathione peroxidase activity in cattle.

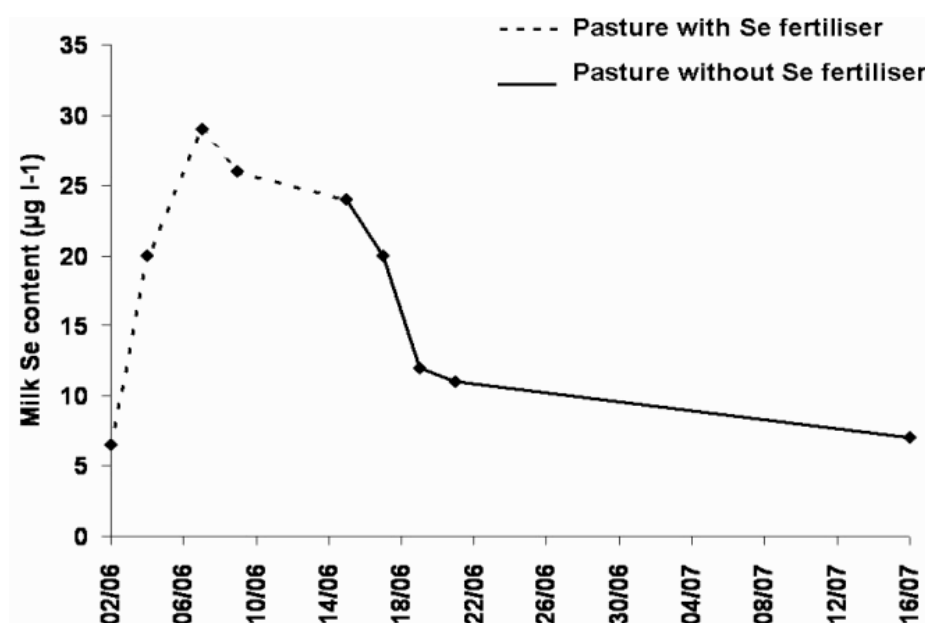


Figure 2. Milk Se content ( $\mu\text{g l}^{-1}$ ).

## Conclusions

The fertilization with Se allowed an increase in Se content in grazed grass, grass and maize silages. The supply of these forages to Belgian Blue cattle lead to improvements in their blood Se status. During the grazing period it was also found to be possible to increase Se milk content. This increase in Se in milk is of interest both for calf and human health.

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# No effects on milk fatty acid composition of perennial ryegrass cultivars with contrasting forage quality

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## Abstract

Plant lipids are a major source of beneficial fatty acids (FA) in milk of ruminants. Forage quality differences can affect rumen metabolism and there could be opportunities to change the composition of ruminant products through species or cultivar choice. Twelve dairy cows were used in two stall-feeding trials with fresh grass to evaluate the effect of perennial ryegrass (*Lolium perenne* L.) cultivars on milk FA composition during the growing season.

Six cultivars were cut daily during three 14-day periods at the same target yield. The experiments consisted of two 3x3 Latin square (LS) trials, in each of which three cultivars were fed to three cows. Each LS was duplicated. The six cultivars differed significantly in concentrations of water-soluble carbohydrates, neutral-detergent fibre and crude protein. However, there were no significant differences among the six cultivars in FA concentration or proportions of FA. No variations in DM intake, milk production and composition or milk FA composition were found. The latter may be due to the lack of variation in grass FA concentration and composition in the cultivars studied.

Keywords: water-soluble carbohydrates, NDF

## Introduction

Plant lipids are a major source of beneficial fatty acids (FA) in milk. Forage quality differences can affect rumen metabolism and there could be opportunities to change the composition of ruminant products through species or cultivar choice (Dewhurst *et al.*, 2006). Therefore, effects of perennial ryegrass cultivars with contrasting forage quality on milk FA composition were evaluated in an indoor feeding experiment.

## Materials and methods

Six diploid perennial ryegrass (*Lolium perenne* L.) cultivars were grown in replicate blocks and cut daily during three 14-day periods between July and August at the same target yield of approximately 2000 kg dry matter (DM) ha<sup>-1</sup>. Samples from day 10 to 13 of each period were pooled and the concentrations of neutral detergent fibre (NDF), ash, acid detergent lignin (ADL), crude protein (CP), and water-soluble carbohydrates (WSC) were determined. At day 11 of each period, a grass sample was collected to determine the FA composition. All cultivars were compared in an analysis of variance for replicate blocks (Genstat 5 release 5.4). Before and after the experiment, and in the two intermittent 14-day periods, the cows were fed two other cultivars, cvs Barlet and Magella (Elgersma *et al.*, 2003).

Twelve multiparous high-producing Holstein Friesian dairy cows in mid lactation (126 ± 21 days in milk) were used. The cows produced 28.8 ± 1.8 kg d<sup>-1</sup> of milk at the start, and 21.9 ± 1.4 kg d<sup>-1</sup> at the end of the experiment. Replicate cows were selected on date of calving. The animals were assigned randomly to the cultivars. The experiments consisted of two 3 x 3 Latin square trials (LS1 and LS2), in each of which three cultivars were fed to three

cows. Each LS was duplicated. The cows were fed fresh grass *ad libitum*, supplemented with 4 kg of concentrates (Tas *et al.*, 2005). The first five days were used for adaptation and the average DM intake (DMI), milk yield (MY) and milk composition (MC) per period was calculated from days 6-13. At each milking, yield was determined and milk sampled. On day 11, a milk sample of 200 ml was stored at -20°C for FA analysis. FA methyl esters were analysed on a Carlo Erba 5200 gas chromatograph, equipped with a flame ionization detector at 280°C.

The LS1 and LS2 were analysed separately. The cow was the experimental unit. Effects of treatment, cow and period on DMI, MY, and milk FA were analysed.

## Results and discussion

Table 1. Herbage yield (HY) (> 5 cm), leaf blade proportion, and concentrations of water-soluble carbohydrates (WSC), crude protein (CP), neutral detergent fibre (NDF), acid detergent lignin (ADL) and fatty acids (FA) of six diploid perennial ryegrass cultivars, mean values from pooled samples taken during days 10-13 during each of three 14-day periods from June-August, standard error of difference, and significance among cultivar means (ns = not significant; \* =  $P < 0.05$ ; \*\* =  $P < 0.01$ ; \*\*\* =  $P < 0.001$ ).

Cultivar	HY (kg DM ha <sup>-1</sup> )	Leaf (% of DM)	WSC (g kg <sup>-1</sup> DM)	CP (g kg <sup>-1</sup> DM)	NDF (g kg <sup>-1</sup> DM)	ADL (g kg <sup>-1</sup> DM)	ASH (g kg <sup>-1</sup> DM)	FA (g kg <sup>-1</sup> DM)
Abergold	2173	82	192	160	399	16.7	110	20.5
Respect	2159	80	152	159	429	18.2	121	21.3
Agri	2149	78	170	157	423	16.5	113	21.0
Herbie	2099	81	172	156	412	17.2	118	21.7
Barezane	1993	86	158	166	414	16.9	119	24.0
Barnhem	2185	88	195	150	400	15.3	116	24.1
Mean	2126	83	173	158	413	16.8	116	22.1
s.e.d.	27	1	5	2	6	1.09	5.4	1.54
Significance	***	***	***	***	**	ns	*	ns

The six cultivars were rather variable in their morphological and chemical characteristics (Table 1). Cvs Barezane and Respect had a lower ( $P < 0.001$ ) WSC concentration than the other cultivars, while Abergold and Barnhem had the highest ( $P < 0.001$ ) WSC and the lowest ( $P < 0.01$ ) NDF concentrations. Barnhem had the lowest, and Barezane the highest CP concentration ( $P < 0.001$ ). Barnhem had a lower and Respect a higher ash concentration than other cultivars ( $P < 0.05$ ). Despite the variation in quality parameters among the cultivars, herbage DMI did not differ, and milk production and composition were similar (Table 2). The similar DMI, MY and MC enabled an unbiased comparison of the effects of grass cultivar quality and composition on milk FA composition in this study. In contrast to some evidence reported in literature (e.g., Miller *et al.*, 2001), the variation in WSC among the cultivars did not affect intake. There was even a tendency for a lower DMI of the high-sugar varieties Abergold and Barnhem (Table 2), as discussed by Tas *et al.* (2005).

In both LS experiments, the concentration and composition of FA in milk did not differ among cows (Table 2), which may be due to the lack of sufficient variation in grass lipid concentration among the cultivars and throughout the season.

In the intermittent 14-d periods, cvs. Barlet and Magella had similar herbage yields and quality, but Barlet consistently had a higher ( $P < 0.001$ ) C18:3 concentration than Magella, and milk fat from cows fed Barlet contained less saturated FA and more CLA ( $P < 0.001$ ) than Magella (Elgersma *et al.*, 2003). This may suggest that variations in lipid composition of the diet would be needed to affect milk lipid composition (intermittent periods), but that plant morphological or chemical characteristics such as ash, NDF, CP, and WSC concentration do

not affect milk FA if the herbage Besides, FA concentration did not differ among cultivars in this study. Further research is needed to test this hypothesis, as there also is evidence that herbage N concentration is positively related to the total FA and C18:3 concentrations (Elgersma *et al.*, 2005; Witkowska *et al.*, 2006).

Table 2. Dry matter intake (DMI), milk yield (MY), milk fat, milk protein, fat-and-protein corrected milk (FPCM), saturated fatty acids (SFA), mono- (MUFA) and poly-unsaturated fatty acids (PUFA) and conjugated linoleic acid (CLA) concentrations in milk of cows in two Latin Square experiments (LS1 and LS2), fed different perennial ryegrass cultivars; mean values from days 11-14 during each of three 14-day periods from June-August, standard error of difference, and significance among cultivar means (ns = not significant; \* =  $P < 0.05$ ).

Cultivar	DMI (kg d <sup>-1</sup> )	MY (kg d <sup>-1</sup> )	Fat (g kg <sup>-1</sup> )	Protein (g kg <sup>-1</sup> )	FPCM (kg d <sup>-1</sup> )	SFA (g kg <sup>-1</sup> fat)	MUFA (g kg <sup>-1</sup> fat)	PUFA (g kg <sup>-1</sup> fat)	CLA (g kg <sup>-1</sup> fat)
<i>LS1</i>									
Abergold	16.2	26.9	40.5	34.3	27.0 <sup>ab</sup>	650	290	39	12.6
Agri	17.4	28.1	39.2	33.7	27.8 <sup>a</sup>	650	289	41	15.3
Barezane	17.4	26.3	38.3	34.1	26.0 <sup>b</sup>	637	297	45	17.4
s.e.m.	0.4	1.2	1.1	0.4	1.2	8	7	4	3.2
Sign	ns	ns	ns	ns	*	ns	ns	ns	ns
<i>LS2</i>									
Respect	16.6	28.2	37.9	32.9	27.5	666	275	40	13.6
Herbie	16.8	27.9	38.5	33.8	27.3	659	279	42	16.2
Barnhem	16.1	26.6	39.7	33.4	26.6	663	277	40	14.5
s.e.m.	0.4	0.9	0.9	0.5	0.8	7	4	3	3.5
Sign	ns	ns	ns	ns	ns	ns	ns	ns	ns

## Conclusions

The six studied perennial ryegrass cultivars differed significantly in morphological and chemical parameters. However, no differences among the six cultivars in FA concentration or proportions of FA were found. DM intake, milk production and milk FA composition were similar when feeding these cultivars to dairy cows. The latter may be due to the lack of variations in grass FA concentration and composition in the cultivars studied.

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# Lamb meat – effect of finishing system on quality

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## Abstract

Norwegian lamb meat production is mainly based on local resources such as natural pastures in forest and mountain areas. Geography and high production costs set limits for Norwegian farmers competing in an international market. Marketing products of unique quality is therefore one strategy to meet international competition. The objective of this study was to examine effects of finishing systems on lamb meat quality. Lambs grazed natural mountain pastures during summer 2006 and 2007 in Troms County, Northern Norway. Four and six weeks prior to slaughtering in 2006 and 2007, respectively, a group of lambs was gathered and let to graze cultivated pastures close to the farm. The rest of the lambs continued grazing the natural pastures. Lambs were slaughtered on the same day and carcass quality and sensory attributes were measured. In 2006, lambs from the cultivated pasture were significantly fatter than lambs from the natural pastures. Some differences in sensory attributes related to pasture composition were found in 2006, but not in 2007.

Keywords: meat quality, sensory attributes, Northern Norway, slaughter quality

## Introduction

Norwegian agricultural policy makers are currently focusing on the potential of increased export of traditional products based on Norwegian natural resources. Quality aspects such as clean nature and animal health are stressed as comparative advantages. This is in line with demands from European consumers who are increasingly focusing on quality aspects of food like origin of raw materials and processing methods, traditional and ethically sound production chains.

Studies have demonstrated a significant relationship between lamb meat quality and type of pasture or feed consumed by the lambs (e.g. Fraser *et al.*, 2004). Duckett (2001) concluded that finishing systems are more important in determining fatty acid composition and flavour in lamb meat than breed of sheep. Most studies have focused on how lamb growth rate and meat quality is affected by different lowland pastures, such as legumes vs. grasses or comparisons of concentrates vs. pasture (Duckett, 2001). In a Norwegian study (Ådnøy *et al.*, 2005) significant differences were found in chemical composition and sensory quality of meat from comparable groups of lambs grazing in unimproved mountain pastures compared with fertilized cultivated lowland pastures during the summer. The objective of the present study was to examine lamb meat quality when lambs were finished on natural mountain pastures.

## Materials and methods

Norwegian white sheep grazed natural mountain pastures (20 km<sup>2</sup> raising 20 to 800 m above sea level) in Troms county in Northern Norway (69°N 18°E) from the middle of June to the end of August in 2006 and to mid-August in 2007. A random group of sheep (40 lambs in 2006 and 39 lambs in 2007) were gathered from the mountain pasture, weighed and put on a



cultivated pasture (60 m<sup>2</sup>) close to the farm. The pasture had a botanical composition common in long term grassland in this region (mainly *Agrostis capillaris*, *Phleum pratense*, *Poa pratensis* and *Deschampsia cespitosa* and herbs such as *Ranunculus repens*, *Taraxacum officinale*, *Rumex crispus*). The remaining lambs (211 in 2006 and 171 in 2007) continued to graze the natural pastures (*Avenella flexuosa*, *A. capillaris*, *Anthoxanthum odoratum* and *D. cespitosa*). In mid-September lambs from both pastures were gathered, weighed (autumn weight) and selected to a live weight above 42 kg. Twenty-two lambs (11 female and 11 male lambs) from each pasture in 2006 and 21 lambs (2 female lambs and 19 male lambs) from each pasture in 2007 were slaughtered. The carcasses were submitted to low voltage electric shock before placed separately on hooks in cool storage at 5°C. After 24 hours in chilled storage the saddles (both loins – *M. longissimus dorsi* – with bones) were removed from the carcasses, vacuum packed and brought to Matforsk AS – Norwegian Food Research Institute. Slaughter data were analysed using Analysis of Variance (Minitab Statistical Software) with pasture as the fixed effect.

After 6 (2006) and 7 days (2007), meat samples were heated to 70°C for 40 min and served from a hot plate at 65°C to a sensory panel consisting of nine selected assessors at Matforsk. The method used was descriptive profiling according to ‘Generic Descriptive Analysis’ (Lawless and Heymann, 1999). Loin slices cut 1.5 cm thick, were evaluated on a scale from 1 (lowest intensity) to 9 (highest intensity) on a total of 19 different sensory attributes. Odour attributes were: sweet, acid, metallic, vernal sweetgrass (*A. odoratum*), leaves/bark and rancid. Flavour attributes were: sweet, acid, bitter, metallic, cloying, vernal sweetgrass, leaves/bark, gamey and rancid. Texture attributes were: hardness, tenderness, fatness and juiciness. Sensory data were analysed separately for each year using a mixed-model ANOVA (PROC GLM, SAS for Windows, Ver 9.1.3 from SAS Institute, Inc., Cary, NC, USA) with the assessor and animal (nested within type of pasture) and their interaction as random effects, and type of pasture as fixed effect. Effect of gender was not significant when included in the model for 2006; consequently we did not include this factor in the current models (for 2006 and 2007). The chosen level of significance was  $P < 0.05$ .

## Results and discussion

Table 1 shows slaughter data of lambs in each pasture group. There were no significant differences in autumn weight, carcass weight and EUROP conformation between pastures.

Table 1. Slaughter data for lamb from different pasture groups in 2006 and 2007.

	2006		SEM	P	2007		SEM	P
	Cultivated pasture	Natural pasture			Cultivated pasture	Natural pasture		
No of lambs	22	22			21	21		
Age at slaughter (days)	132	133			128	130		
Autumn weight (kg)	47.4	46.2	1.0	0.37	48.0	48.7	1.3	0.72
Carcass weight (kg)	19.8	19.9	0.5	0.81	19.6	21.3	0.6	0.053
EUROP Conformation <sup>1</sup>	8.0	7.6	0.3	0.49	8.6	9.3	0.3	0.09
Fatness <sup>2</sup>	5.5	4.8	0.2	0.04	4.8	4.9	0.2	0.76

<sup>1</sup>EUROP Conformation: P- = 1, P = 2, P+ = 3, ... R- = 7, R = 8, R+ = 9.

<sup>2</sup>Fatness: 1- = 1, 1 = 2, 1+ = 3, 2- = 4, 2 = 5, 2+ = 6.

Lambs grazing cultivated pasture were significantly fatter than lambs grazing natural pastures in 2006 ( $P < 0.05$ ). Lambs grazing the cultivated pasture were less active and thus more likely to gain more fat during the grazing season. A difference in fatness would therefore have been expected also in 2007.

Table 2 shows average values of sensory attributes for which significant differences between the two pasture groups were found in 2006 or in 2007. In 2006, meat from lamb grazing on natural pastures had a lower intensity of rancid flavour, was less hard, tenderer and less fatty than meat from lamb grazing on cultivated pasture. The only significant effect found in 2007 was for juiciness, which was highest in meat from lambs grazing natural pastures. This shows that the effects of finishing lambs on natural pastures compared with the diverse cultivated pasture were rather small under the prevailing conditions. The reason for the incompatible results between the two years may either be climatic differences between years or other unrevealed factors.

Table 2. Average values of sensory attributes for which significant differences between the two pasture groups were found in 2006 or in 2007.

	2006			2007		
	Cultivated pasture	Natural pasture	<i>P</i>	Cultivated pasture	Natural pasture	<i>P</i>
No of lambs	22	22		21	21	
Rancid flavour	1.21	1.03	0.03	1.13	1.17	0.61
Hardness	3.72	3.18	< 0.01	3.66	3.54	0.43
Tenderness	5.68	6.62	< 0.01	6.28	6.49	0.42
Fatness	3.96	3.63	< 0.01	4.47	4.52	0.52
Juiciness	5.41	5.19	0.06	6.01	6.27	< 0.01

## Conclusions

Finishing system had small effects on meat quality in this study, and either climatic differences between years or other unrevealed factors affected the results. More systematic experimental research is needed in this field to explore further effects of different pasture systems on lamb meat quality.

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## Effect of different grazing times on milk fatty acid composition

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### Abstract

An experiment was performed with 15 cows in a 3 x 3 Latin Square design. Animals on the three treatments received a total mixed ration (TMR) *ad libitum* combined with no grazing (TMR00), with grazing 6 h (TMR06) or with grazing 12 h (TMR12) after morning milking. The purpose was to determine the influence of different grazing times on milk fat profiles. The results showed that TMR intake was 20, 16 and 12 kg DM cow d<sup>-1</sup> for TMR00, TMR06 and TMR12 respectively. Milk yield was affected by treatments, averaging 34 kg d<sup>-1</sup> in TMR00 and TMR06, while on TMR12 it was 32 kg d<sup>-1</sup> ( $P < 0.05$ ). Milk composition was also affected, except milk protein concentration (30.4 g kg<sup>-1</sup>). In this sense, milk lactose was higher for TMR00 (50.97 g kg<sup>-1</sup>,  $P < 0.05$ ) compared with TMR06 or TMR12 (49.81 and 50.73 g kg<sup>-1</sup>). Milk fat was lower in TMR12 (36.2 g kg<sup>-1</sup>,  $P < 0.05$ ) than TMR00 (38.2 g kg<sup>-1</sup>) and TMR06 (38.8 g kg<sup>-1</sup>). In relation to the milk fatty acid profile, cows grazing had higher proportion of *cis*9, *trans*11 CLA and C18:1t11 compared with those feeding only TMR.

Keywords: Milk fatty acids, CLA, grazing times, total mixed ration, milk quality

### Introduction

Fat is the most variable component of milk, and its synthesis is affected by several nutritive and environmental factors (Bauman *et al.*, 2001). Nowadays, the milk fat is being criticised, because in the European diet around 50% of saturated fat comes from milk origin. However, about 2% of total fatty acids in milk fat are polyunsaturated. In recent years, several works have studied the role of some fatty acids, especially the conjugated linoleic acid (CLA), with specific biological actions, as potential anti-carcinogen, anti-atherogenic, anti-diabetic, and anti-obesity in human health benefits (Bauman *et al.*, 2006). The CLA is a mix of positional and geometric isomers of linoleic acid, being the C18:2 c9 t11 the main isomer. These positive findings have led to interest in dairy cow nutrition as a way to modify the fatty acid profile of milk fat through dietary manipulation. It is well established that cows fed with only pasture generally have higher milk CLA concentrations (Khanal *et al.*, 2005), specifically the C18:2 c9 t11 isomer, than with indoor feeding. However pasture-based systems for high producing dairy cows need supplementary feeds to increase nutrient intake in order to support high milk production. The knowledge of feeding mixed systems of pasture-total mixed ration (TMR) on profile fatty acids is limited. There is a strong relationship between milk fatty acid composition and feeding strategy. In this sense, supplementing pasture with TMR could have the advantage of increased milk yield with enhanced fat and protein contents. The objective of this work was to compare the effects of different grazing times associated with fed TMR on milk fatty acid profiles.

### Materials and methods

Fifteen Holstein dairy cows (610 ± 45 kg liveweight; 34.8 ± 6.8 L d<sup>-1</sup>) in the first third of lactation were blocked into three groups and randomly assigned to three treatments: 1) TMR00, feeding only with TMR; 2) TMR06, 06h grazing plus TMR; 3) TMR12, 12h grazing plus TMR. The TMR was formulated for a milk production of 30 l d<sup>-1</sup> including

maize and legume silages, cereal straw and concentrate and offered *ad libitum*. Cows with production higher than 30 l were supplemented with concentrates. Cows in TMR06 and TMR12 groups were moved to a fresh paddock after morning milking. After an adaptation period of fourteen days, the TMR intake and milk production were recorded daily during the assay period (seven days). Daily, the TMR (offered and refused) and milk (in both milkings) were sampled. The pasture was sampled the first and last days in each assay. Feedstuffs and pasture were analysed for dry matter (DM), organic matter (OM), crude protein (CP) and neutral and acid detergent fibre (NDF, ADF) by near infrared reflectance spectroscopy (NIRS). Milk samples were analysed by MilkoScan FT6000. Milk fatty acids were extracted and analysed after derivatisation using the methodology described by Sukhija and Palmquist (1988) by gas chromatography (Varian Star 3400 CX). Statistical analysis was performed in SAS (1999) using the GLM procedure for a design 3 x 3 Latin square.

## Results and discussion

Pasture and TMR chemical composition are showed in Table 1. Production parameters from cows on three feeding systems based in grazing times with TMR showed that dry matter intake (DMI), including TMR and extra concentrate, was highest for TMR00 (23.3 kg d<sup>-1</sup>), lowest for TMR12 (14.9 kg d<sup>-1</sup>) and intermediate for TMR06 (18.9 kg d<sup>-1</sup>) (Table 2). The lower dry matter intake in TMR12 group resulted in the lowest milk production. However, there were not differences in milk production between TMR00 and TMR06 groups in spite of the lower intake in the TMR06 pasture system. Milk protein yield was not affected by pasture system; nevertheless the milk fat content was higher for TMR00 and TMR06 (38.2 and 38.8 g kg<sup>-1</sup>) than TMR12 (36.2 g kg<sup>-1</sup>).

Table 1. Chemical composition (mean  $\pm$  standard error) of forage and total mixed ration (TMR) during experimental period.

	TMR	Forage
DM (g kg <sup>-1</sup> )	508.7 $\pm$ 14.0	157.8 $\pm$ 12.0
OM (g kg <sup>-1</sup> DM)	907.6 $\pm$ 9.0	894.0 $\pm$ 6.0
CP (g kg <sup>-1</sup> DM)	151.7 $\pm$ 7.0	198.0 $\pm$ 34.0
NDF (g kg <sup>-1</sup> DM)	401.9 $\pm$ 19.0	387.6 $\pm$ 60.0
ADF (g kg <sup>-1</sup> DM)	243.9 $\pm$ 26.0	197.6 $\pm$ 52.0

Table 2. Dry matter intake (DMI), milk production and composition under different dairy cows feeding strategies.

	Feeding System			MSE	P <sup>1</sup>
	TMR00	TMR06	TMR12		
DMI TMR (kg d <sup>-1</sup> )	20.23 <sup>c</sup>	15.85 <sup>b</sup>	11.79 <sup>a</sup>	0.267	*
Milk yield and composition					
Milk yield (kg d <sup>-1</sup> )	33.55 <sup>ab</sup>	34.33 <sup>b</sup>	32.33 <sup>a</sup>	0.349	*
Milk protein (g kg <sup>-1</sup> )	30.10	30.88	30.90	0.091	ns
Milk fat (g kg <sup>-1</sup> )	38.22 <sup>b</sup>	38.80 <sup>b</sup>	36.23 <sup>a</sup>	0.266	*
Milk lactose (g kg <sup>-1</sup> )	50.97	49.81	50.73	0.186	ns

<sup>1</sup>Significative statistical probabilities \*P < 0.05; ns: Non Significant; TMR: Total Mixed Rations.

The fatty acid composition of milk fat is presented in Table 3. The TMR12 system resulted in a lower concentration of medium-chain fatty acids, while the long-chain fatty acids are increased with the pasture intake. The C18:3 proportion increased as a result of increasing grass intake, but significant differences were not reached.

The linoleic and linolenic acids are biohydrogenated into the rumen until stearic acid with trans-vaccenic acid as an intermediate product. Both are absorbed and could be desaturated in the mammary gland to oleic and rumenic acids respectively. In milk, there are no differences

between treatments in the concentration of C18:0, while the proportion of trans-vaccenic acid increased significantly with TMR12 treatment. Over both fatty acids, via the enzyme delta-9 desaturase, there are endogenous syntheses of C18:1t11 and C18:2c9t11; however, there were only statistical differences between treatments in trans-vaccenic acid content.

Table 3. Fatty acids (g 100g<sup>-1</sup> FA) profile in milk fat under different dairy cows feeding strategies.

	Feeding System			MSE	P <sup>1</sup>
	TMR00	TMR06	TMR12		
Short-chain fatty acids	15.38	13.55	17.24	3.045	ns
Medium-chain fatty acids	54.67	52.98	44.46	3.522	ns
C18:0	14.20	14.81	18.40	1.310	ns
C18:1, <i>cis</i> 9	1.28	1.58	0.66	0.73	ns
C18:1, <i>trans</i> 11	1.37 <sup>a</sup>	1.73 <sup>a</sup>	2.82 <sup>b</sup>	0.294	*
C18:2, <i>cis</i> 9 <i>trans</i> 11	1.92	1.84	1.37	0.055	ns
C18:2, <i>trans</i> 9 <i>trans</i> 12	7.33	7.76	11.18	0.849	ns
C18:3, <i>cis</i> 9 <i>cis</i> 12 <i>cis</i> 15	0.33	0.39	1.25	1.35	ns
Others Long-chain fatty acids	3.83	6.16	4.72	0.832	ns

<sup>1</sup>Significative statistical probabilities \*P < 0.05; ns: Non Significant; TMR: Total Mixed Rations

## Conclusions

The results of this work show that it is possible to change the profile of fatty acids in fat milk with grass intake complementary to a total mixed ration, resulting in an increase in the trans-vaccenic acid content in milk fat.

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# Effects of vegetation and grazing preferences on the quality of alpine dairy products

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## Abstract

This study focuses on alpine outfields and summer farms in Norway. The main objective is to investigate relationships among vegetation composition, grazing preferences and quality of dairy products. A broad range of methods to document cattle area use and grazing preferences are used. The study started in 2007 and the results of standard milk analysis are presented here. The standard analyses of the milk on two summer farms showed generally normal and good values of protein content, fat content and the flavour of the milk. To find out if the milk from summer farms has better qualities than other milk it will be necessary to make further studies on the fatty acid composition and other quality measures of the milk. Results regarding milk yield, milk composition and flavour were similar between the two farms despite higher contents of protein and fat in the concentrate given to the cows on one farm. The results underline the need for further studies on the vegetation composition of the pastures and the grazing preferences to explain the differences between the two summer farms and to find connections between important grazing plants and quality properties of the milk.

Keywords: alpine pastures, grazing preferences, milk quality, aerial photos, GPS, GIS

## Introduction

In Norway summer farming has created unique cultural landscapes now used as a basis for development of ecotourism and local food products. However, summer farming has declined considerably during the 20th century, but in the regions of Valdres (Oppland County) and Hallingdal (Buskerud County) summer farming (transhumance) is still practised. Here most dairy farms produce milk in the mountains for a period of about 8 weeks in summer. This production system based on free-range grazing has recently been studied with regard to economy and biodiversity in a project called 'Living Summer Farms' (Tuv, 2002). This project showed that some adjustments of the production system could improve the economic results significantly, while the production still contributes to the maintenance of the summer farming landscape and biodiversity (Tuv, 2002). In 2004 the Norwegian University of Life Sciences investigated the effect of grazing on milk quality in different regions. Preliminary results show variations among districts in fatty acid composition, but so far the reasons for the variation have not been identified (Harstad, pers. comm.). Other preliminary studies indicate that milk products from summer farms may have a higher content of unsaturated fatty acids, considered as beneficial for human health (Wetlesen, pers. comm.). Also, other studies indicate that dairy products based mainly on outfield grazing may have other qualities than conventional dairy products. If these indications can be better documented it may increase the market value of such products and strengthen the summer farming economy. Against this background a PhD-study started in 2007 as part of a more comprehensive project 'Farming

systems for high quality food products and sustainable agriculture in mountainous areas'. The objective of the PhD-study is to investigate effects of vegetation composition and grazing preferences on the quality of summer farm dairy products. Here we present preliminary results from standard milk analysis and other standard data from the summer season in 2007.

## Materials and methods

The investigations are carried out at two summer farms, Buodden in Valdres and Hamarsbøen in Hallingdal, with outfields that have been used for grazing since a long time ago (Lunnan *et al.*, 1999). These two summer farms were chosen because of their quite similar and extensive production systems. In 2007 they had 12 milking cows at Buodden and 18 at Hamarsbøen. Buodden is situated in the north boreal vegetation zone, 950 m a.s.l. Hamarsbøen is situated 1040 m a.s.l. in the transition between the north boreal and low alpine vegetation zones. On both study sites the bedrocks have a high weathering capacity giving rise to soils of intermediate or good nutritional quality for plants. The cattle stay at the summer farms from the beginning of July to the beginning of September. The area use and grazing preferences of the dairy cattle in the outfields of the two summer farms are investigated by GPS studies of the cattle combined with aerial photo interpretation of the vegetation, field observations and microhistological analysis of faecal samples. The composition of the grazed vegetation is also documented by pin-point and plot analysis, and the nutrient content of important grazing plant species analysed. In 2007 milk samples were taken from 9-10 individuals in each herd, once in July and once in August, on both study sites. All the individuals belong to the breed Norwegian Red (NR) and calved during the winter/early spring (except for 3 cows at Hamarsbøen which calved in October 2006). Calving during winter/early spring gives higher milk yields on the summer farm. Milk amounts (kg), concentrate (type and amounts), calving date and health history for each of the cows is recorded. In addition to standard quality parameters of fat content, protein content, flavour and urea levels, properties such as fatty acid composition, colour, vitamin content (A, E) and oxidation stability of the milk will be studied. For the sensory evaluation of the flavour of the milk a hedonic scale from 1 to 5, with 5 as best score, was used. Products like sour cream, butter and cheese, where grazing effects on the milk properties could be more distinct, will be produced and examined.

## Results and discussion

The results of the milk analysis in 2007 are presented in Table 1. Mean values of the protein content were good on both farms. For milk with a protein content  $< 32 \text{ g kg}^{-1}$  the farmers will be paid less according to the milk payment system in Norway. A normal fat content is 39-42  $\text{g kg}^{-1}$ . At Buodden the fat content was quite high in June, and normal in July and August. At Hamarsbøen the fat content was normal all three months but it increased from June to July. In 2007 the cattle at Buodden and Hamarsbøen received on average 5.6 and 5.9 kg concentrate, respectively, in July, and 4.6 and 5.8 kg, respectively, in August. At Buodden the concentrate consisted of 189 g crude protein and 48 g fat per kg; at Hamarsbøen 151 g and 38 g. It is expected that the milk production declines on the summer farms since the cows are given less concentrates. In 2007 the lactation yields were generally higher at Hamarsbøen than at Buodden. The decline from July to August on both farms was probably because some individuals were in a late lactation stage. Almost all individuals received score 5 for flavours in milk, which is the highest score. None of the cows was treated for illnesses during the summer 2007. The urea content should ideally be between 3 and 6  $\text{mmol l}^{-1}$ . The urea level in August 2007 was therefore low on both farms. Falling urea levels during outfield grazing are common and documented in the project 'Living summer farms' (Tuv, 2002). Low urea values mean that most of the crude protein (CP) is degraded and captured by the rumen microbes.



This process requires adequate amounts of fermentable carbohydrates (CHO). Low urea levels are therefore an expression of too little CP in relation to the amounts of CHO in the rumen. This can cause reduced milk yield and milk quality and sometimes give animal health problems. In 'Living summer farms' the urea levels were increased by 1 mmol l<sup>-1</sup> when the cows were given concentrates with high contents of crude protein. In 2007 the animals at Buodden were given concentrate to compensate for expected low protein levels in the coarse fodder. The cows at Hamarsbøen, however, did not get such high protein concentrate. Still the urea values were similar at the two farms. The reason may be that there is a better balance between CP and CHO in the coarse fodder at Hamarsbøen. There is a common understanding in Norway that milk produced on summer farms has a higher fat content than other milk. The results from Hamarsbøen strengthen this common understanding. Interestingly, the milk fat content recorded in July equalled that at Buodden even though the cows at Buodden were given a concentrate with a higher fat content. Fat in the diet produces fat in the milk, but degradation of fibre in rumen gives milk fat as well. Too much dietary fat decreases fibre digestibility and milk fat. The fat is important for the taste of the milk and the fatty acid composition may be important from a health perspective. It is therefore of importance for the development of branded summer-farm products to analyse the fatty acid composition at the summer farms and to find possible connections among the fatty acid composition, available vegetation types and grazing preferences.

Table 1. Mean values and standard deviations (SD) of the analysis of flavour (scale 1-5 where 5 is best), fat, protein and urea contents of milk, milk yield and concentrate amounts given to cows on the summer farms. At Buodden n = 10 in June and July and 9 in August, at Hamarsbøen n = 9 in June and 10 in July and August.

	Fat* (g kg <sup>-1</sup> )			Protein (g kg <sup>-1</sup> )		Flavour (1-5)		Urea (mmol/l)		Milk (kg)			Concentrates (kg)		
	June	Jul	Aug	Jul	Aug	Jul	Aug	June	Aug	June	Jul	Aug	June	Jul	Aug
Buodden	44	42	41	34	34	4.7	4.6	6.5	2.6	22.6	17.0	15.4	6.1	5.6	4.6
1 SD	2	0	2	2	2	0.7	0.7	0.8	0.4	3.4	2.3	2.6	0.7	0.6	1.5
Hamarsbøen	39	42	39	34	34	5.0	4.7	5.2	2.8	27.0	23.0	22.1	6.5	5.9	5.8
1 SD	2	1	0	3	3	0.0	0.7	0.7	0.5	4.4	4.0	4.3	1.0	0.3	0.4

\*Measures of fat content in milk from individual cows showed unreliable high values probably due to an error in sampling methods. Fat content is therefore based on samples from the milk collecting tank at the two farms. Milk from milk collecting tank was analysed twice during the month; in the middle and at the end of the month.

## Conclusions

Further studies are needed on the fatty acid composition and other quality measures of the milk from summer farms compared with other milk. Previous studies have shown that cows have certain grazing preferences and use the landscape and the vegetation types in a conscious way (Sickel and Norderhaug, 2001; Sickel *et al.*, 2004). Studies of vegetation composition, grazing preferences and plant nutrient contents are needed to explain the differences between the two summer farms reported here, in terms of milk quality.

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# Somatic cell count and quality of milk during pasture turnout of dairy cows

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## Abstract

The aim of the study was to investigate whether spontaneous increase in somatic cell count (SCC) in response to pasture let-out has an effect on milk quality. A total of 35 dairy cows were included. The cows were machine milked twice a day and milk samples were collected for 7 days prior to, and for 5 days following, pasture turnout for analyses of fat, protein, lactose, SCC and Free Fatty Acids. For statistical evaluation of the difference between the values (mean) before the pasture let-out and the values at each sampling occasion thereafter, repeated measures of analysis of variance was calculated using the Mixed Model procedure of SAS. There was a marked peak in milk SCC in response to pasture turnout. However, the milk quality was not affected by the short SCC peak.

Keywords: grazing, somatic cell count, SCC, milk quality

## Introduction

Milk somatic cell count (SCC) is a predominant criterion for milk quality and for the price paid to the farmer because it has been observed that high SCC decreases the milk quality. It is therefore important to increase the knowledge of the causes of variation of milk SCC. The SCC is known to increase during the grazing season but the question is, does this increase affect the milk quality? The aim of the study was to investigate if there is a spontaneous increase in SCC when cows are let out on pasture, and if so whether it has an effect on milk quality.

## Materials and methods

A total of 35 dairy cows were included in the study carried out at Kungsängen Research Centre, SLU, Uppsala. The cows were in lactation day 12-442, lactation no 1-5 and yielding 14.9-39.7 kg Energy Corrected Milk (ECM) at the start of the experiment. They were held in individual tie stalls with straw and sawdust bedding. The cows were individually fed silage and concentrate four times per day prior to pasture turnout and two times per day in connection with milking when they had access to the pasture area. The cows were machine milked twice a day, at 06.30 and 16.00. Sampling was conducted during seven days prior to, and during five days after pasture turnout. Milk samples were collected for analyses of the milk composition (fat, protein, lactose) and SCC. For the afternoon milking, samples were also collected for analyses of Free Fatty Acids (FFA) and casein. The data were analysed using the Mixed Model procedure with repeated measures in SAS 9.1 (SAS, 2002).

## Results and discussion

The SCC increased ( $P < 0.001$ ) all days following pasture turnout for both morning and afternoon milking. There was a marked peak during the afternoon milking the same day as pasture turnout (Day 0). A peak was also seen during the morning milking Day 1 (Figure 1).

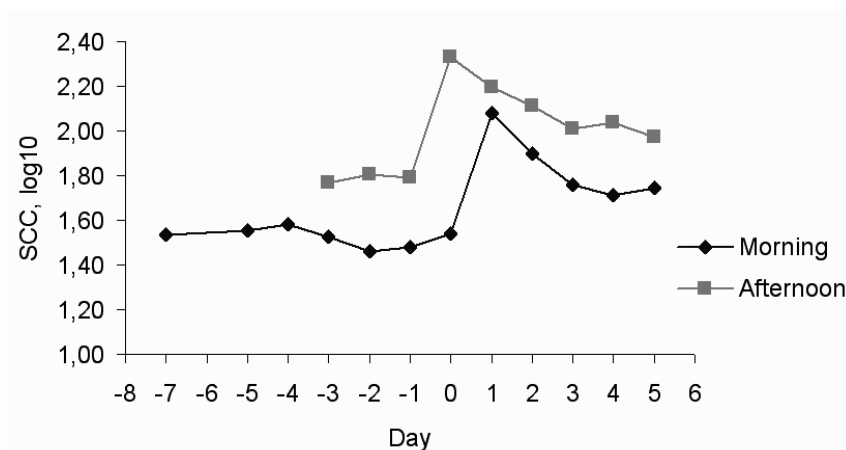


Figure 1. The SCC (log<sub>10</sub>/ml) during morning and afternoon milking. The cows were turned out on pasture following the morning milking Day 0.

The experiment confirms that there is an increase in milk SCC in response to pasture turnout, which is in contrast to what was observed by Pomès *et al.* (2000). However, the cows in our experiment had very low milk SCC prior to pasture turnout and the baseline values of SCC ranged between 5,000 and 222,000 cells ml<sup>-1</sup> during the morning milking, and between 15,000 and 433,000 cells ml<sup>-1</sup> during the afternoon milking. Although mastitis, caused by bacterial infection, is the main causative agent for elevated SCC in milk (Brolund, 1985) it is questionable if the peak we observed was due to infection. Even a slightly increased SCC is associated with a decreased milk yield (Miller *et al.*, 1983). There was a significant drop in milk yield during the morning milking Day 1 (13.7 kg ECM) compared with the baseline value (14.9 kg ECM) but no drop during the afternoon milking. From Day 3, the milk yield instead increased significantly during both morning and afternoon milking compared with the baseline value. The concentration of fat increased ( $P < 0.001$ ) all days compared with the baseline value whereas for the afternoon, fat decreased Day 0 and then increased Days 2 and 3 to a level similar to the baseline (Figures 2 and 3). The FFA level was significantly lower during the afternoon milking Day 1 (0.65 mEq l<sup>-1</sup>) compared with Day -1 (0.98 mEq l<sup>-1</sup>). The decreased fat content during the afternoon milking Day 0 could be an effect of inefficient milk removal in response to the pasture turnout whereby the increased fat content morning milking could be a carry over effect from the preceding milking. Since the FFA value decreased Day 1 compared with Day -1, it is likely that neither the SCC peak nor the pasture turnout impaired the milk fat quality.

In accordance to Miller *et al.* (1983) the percentage of protein increased with increased SCC. They found an increased protein and fat content with increased SCC and no bacteria present. In the present experiment the protein increased ( $P < 0.001$ ) all days compared with the baseline value during the morning milking but decreased Day 0 and Day 1 during the afternoon milking (Figures 2 and 3). The casein level increased ( $P < 0.001$ ) Day 3 and Day 5 compared with Day -1 whereas whey did not change due to pasture let-out. This result indicates that the SCC peak was not a response to infected udders.

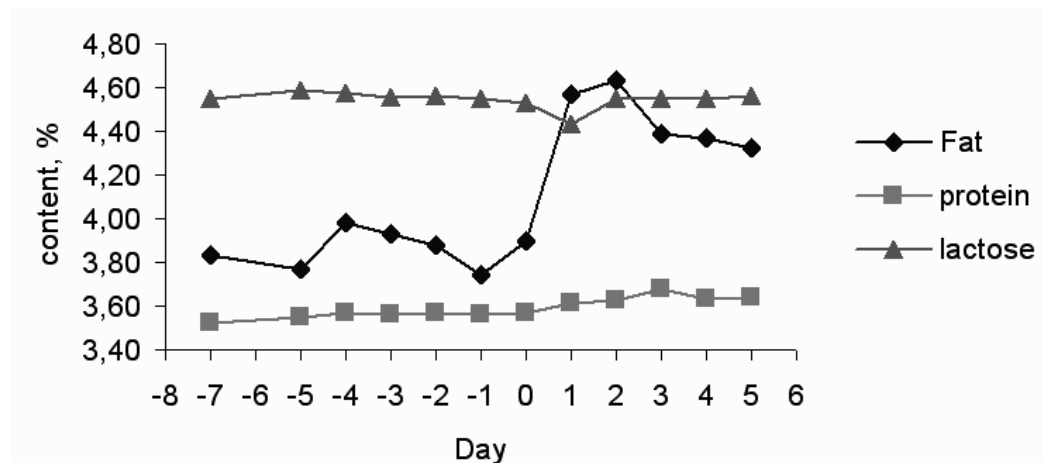


Figure 2. Content (%) of fat, protein and lactose during the morning milking. Following morning milking Day 0 the cows were turned out on pasture.

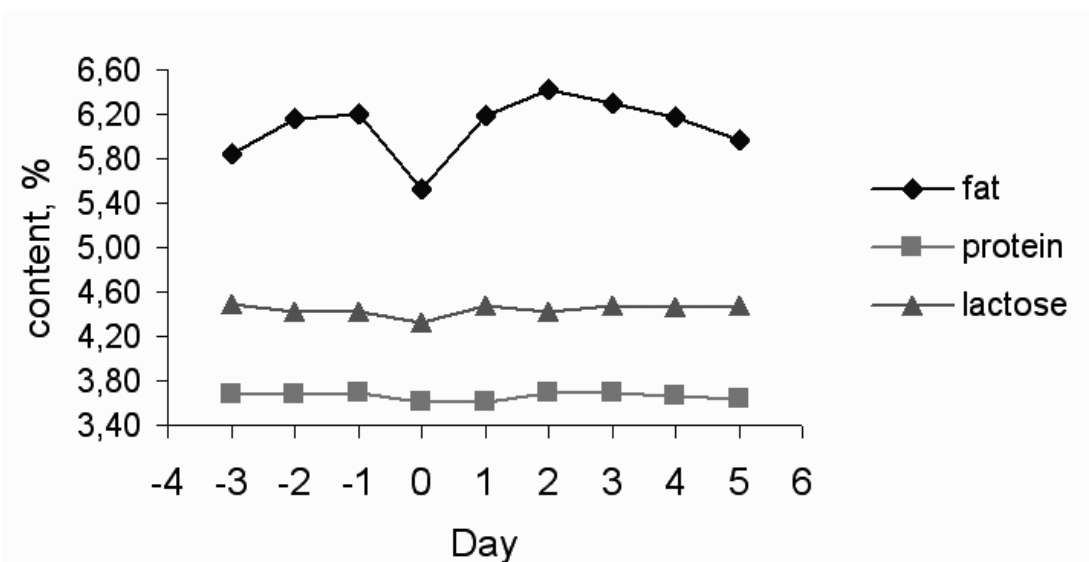


Figure 3. Content (%) of fat, protein and lactose during the afternoon milking. The cows were out on pasture from Day 0.

## Conclusions

In conclusion, there was a marked peak in milk SCC in response to pasture turnout. However, the milk quality was not affected by the short SCC peak and one explanation could be the fairly low level of SCC even with a marked peak.

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# Influence of forage from grassland on the fatty acid content of milk fat

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## Abstract

A trial was carried out to investigate the influence of forage from a temporary grassland and a permanent grassland, and of the addition of maize silage, on milk fat composition. The trial lasted for seven weeks: a two-week adaptation period and five weeks for the trial itself. The forage was given *ad libitum* in the stable. All cows received no concentrate, only a mineral supplement was added.

The feed intake and milk production were recorded daily. Before the trial, and three times during the trial, milk samples were taken for analysis of fat, protein and lactose contents, and also of the different fatty acids in the milk fat. The forage from the temporary grassland contained more than 85% of grasses. In the permanent grassland, only 45% grasses and about 45% of herbs (dandelion) were found in the young forage.

Milk from cows in the variant with permanent grassland had, in comparison with the forage from the temporary grassland, higher proportions of unsaturated fatty acids and higher contents of omega-3 and of conjugated linoleic acid (CLA). With increasing age of the forage, the contents of omega-3 and CLA decreased. The addition of maize silage had a stronger influence on these fatty acids, and the lowest amounts of omega-3 and CLA were found with this variant.

Keywords: permanent and temporary grassland, fatty acid content, CLA, Omega-3

## Introduction

Milk composition, in terms of components considered beneficial to human health, can be directly influenced by dairy cattle feed. This applies in particular to certain fatty acids like omega-3 and conjugated linoleic acids (CLA). Studies carried out by Morel *et al.* (2005; 2006) and van Dorland (2006) showed that the type of a sown grass mixture (grasses or legumes) influences the fatty acids in the milk. This study aims to assess the influence of forage from temporary or permanent grassland, and of the addition of maize silage, on the milk fat composition.

## Materials and methods

The following three variants were investigated:

Variant 1: forage from a temporary grassland

Variant 2: forage from a permanent grassland

Variant 3: forage from a permanent grassland and maize silage

Each variant was carried out with six dairy cows. After a two-week adaptation period with the same feeding regime of pasture and hay, the trial with the three variants was carried out for five weeks. The forage from the temporary and permanent grassland was cut daily and given *ad libitum* in the stable. During the adaptation period and the three first weeks of the trial, forage from the first cut was fed. In the fourth and fifth week the forage was from the second cut. In variant 3 the cows received in addition 5 kg dry matter of maize silage. The cows received no concentrates, only a mineral supplement was added. Feed intake and milk production were recorded daily.

Every day a sample was taken from the grass to determine the dry matter content. The samples were pooled and once a week the nutrient contents were analysed. The maize silage was also analysed once a week. The mineral supplement was analysed two times during the trial. After the adaptation period, and three times during the trial milk, samples were taken for analysis, and in addition to the fat, protein and lactose contents, the different fatty acids in the milk fat were analysed. The milk fat composition was analysed after Collomb and Bühler (2000). Data were statistically analysed using an analysis of variance and a Newman-Keuls test.

## Results and discussion

Forage from the temporary grassland contained more than 85% grasses, the rest was mainly clover. In the permanent grassland, grasses comprised only 45% in the young forage, and herbs (mainly *Taraxacum officinale*) also comprised about 45%. With increasing age of the forage the proportion of grasses increased to 70%, and the proportion of herbs decreased to 15%. In both the temporary and permanent grassland, linolenic acid dominated with a proportion of > 60% of the forage fat content. The different fatty acids in the grass and maize silage are shown in Figure 1. The fatty acid composition of the fat in the forage in this investigation was similar to the values obtained by Morand-Fehr and Tran (2001).

The maize silage had an average dry matter content of 37%, with 31 g ash, 76 g crude protein and 196 g crude fibre per kg DM.

The average amount of energy-corrected milk at the end of the adaptation period was 27.2, 26.0 and 27.4 kg for the three variants. During the five weeks the milk performance decreased to 23.2, 22.6 and 21.5 kg. The fat content in the milk was 4.2, 4.1 and 4.1%, and increased in all three variants during the trial to 4.3, 4.4 and 4.6%.

The protein content was 3.7, 3.6 and 3.5% at the end of the adaptation period, and during the trial it decreased to 3.5, 3.5 and 3.2%. In variant 3, the content was significantly lower in comparison with the other two variants.

In variant 2 with forage from the permanent grassland the amount of saturated fatty acids in the milk fat was lower in comparison with the variant 1 (forage from temporary grassland) and variant 3 (with maize silage). Variant 2 had the highest amounts of mono- and polyunsaturated fatty acids, and in comparison with the forage from the temporary grassland, higher contents of omega-3 (Figure 2). The addition of maize silage also reduced the omega-3 fatty acids.

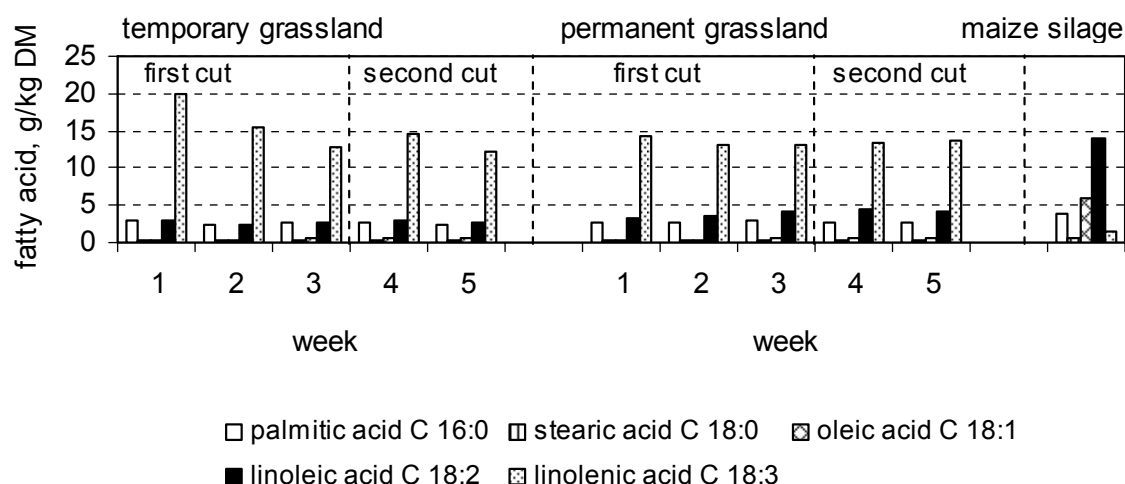


Figure 1. Fatty acid composition of the fat in the forage from the temporary and permanent grassland and maize silage.

A similar effect was found with CLA. Variant 2 also had the highest amounts and the addition of maize silage resulted in the lowest amounts of CLA being found (Figure 3). Furthermore, with increasing age of the first cut forage, the contents of omega-3 and CLA also decreased.

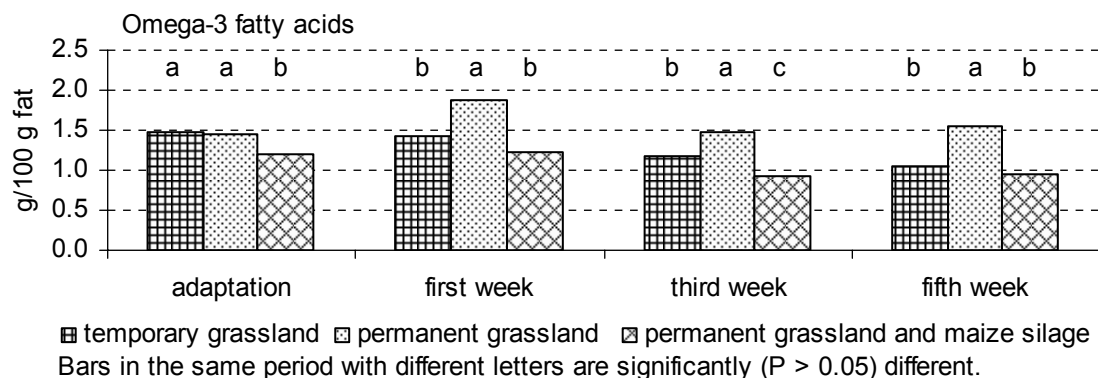


Figure 2. Development of the omega-3 fatty acids in the milk fat of the three variants.

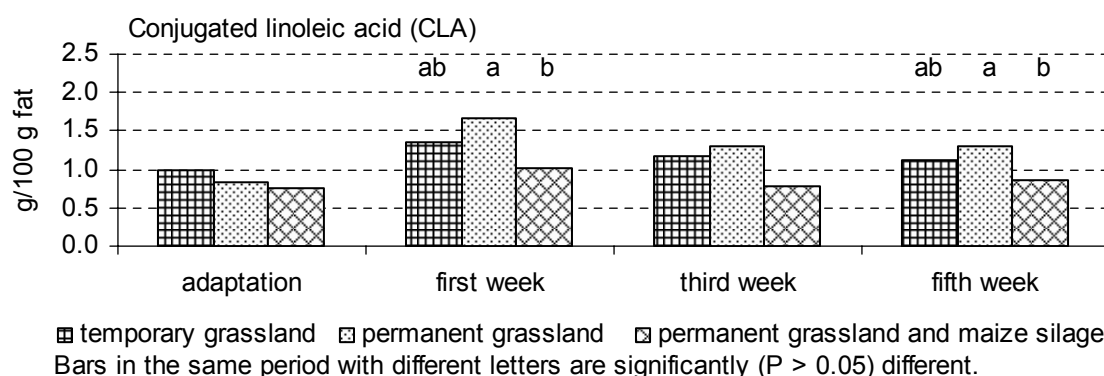


Figure 3. Development of the conjugated linoleic acid in the milk fat of the three variants.

## Conclusions

- Fat of forage from a temporary and from a permanent grassland contained more than 60% of linolenic acid.
- The milk of cows fed with forage from the permanent grassland had, in comparison with the forage from the temporary grassland, higher contents of omega-3 and of conjugated linoleic acid.
- In the variant with the addition of maize silage the lowest contents of omega-3 and CLA were found.

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## **Session 2B**

### **Feed quality**

# Estimation of nutritive value of grasses from semi-natural grasslands by biological, chemical and enzymatic methods

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## Abstract

The nutritive value of seven native temperate grass species from semi-natural grasslands (*Lolium perenne*, *Poa trivialis*, *Festuca rubra*, *Dactylis glomerata*, *Anthoxanthum odoratum*, *Elymus repens*, *Arrhenatherum elatius*) was investigated. The species were cultivated in monocultures in a fully factorial design with 3 replicate blocks, 2 levels of nitrogen supply (120 and 360 kg N ha<sup>-1</sup> year<sup>-1</sup>) and 6 cuts per year. Samples were harvested at the first cut in May, and nutritive value assessed by chemical composition, estimation of organic matter digestibility (OMD) by pepsin-cellulase digestibility, and by the measurement of the dry matter (DegDM) and crude protein (DegN) degradability in the rumen of cows using the nylon bag technique. Effect of N fertilization was not significant on OMD, DegDM and DegN. There were significant differences ( $P < 0.05$ ) between species for DegDM and DegN with highest values for *L. perenne*, *P. trivialis*, *A. odoratum*, and the lowest value of DegDM for *F. rubra* and of DegN for *D. glomerata*. DegDM and OMD values were closely related ( $R^2 = 0.91$ ). The low DegN of *D. glomerata* may contribute to improved protein value.

Keywords: native grass species, nutritive value, digestibility, rumen degradability, nitrogen fertilization

## Introduction

In semi-natural temperate grasslands, perennial C3 grasses are the most abundant plants (Louault *et al.*, 2005). However, most of these species are not used as cultivated grass crops and there are few estimates of their nutritive value for ruminants. When *in-vivo* measurements are not possible, estimation of nutritive value can rely on the analysis of the chemical composition, on enzymatic digestibility and on rumen *in situ* degradability measurements. The aim of this study was to characterize the nutritive value of seven perennial C3 native grass species found in semi-natural grasslands in central France (Auvergne), with a particular focus on the rumen degradability of dry matter (DegDM) and nitrogen (DegN) in relation to chemical composition and to digestibility estimates.

## Materials and methods

Seven perennial C3 grasses that co-occur in semi-natural grasslands were studied: *Lolium perenne*, *Poa trivialis*, *Festuca rubra*, *Dactylis glomerata*, *Anthoxanthum odoratum*, *Elymus repens* and *Arrhenatherum elatius*. These species are among the 20 most widely distributed *Poaceae* species in the French Massif Central. Seeds of these grass species were collected in their native habitat. They were sown in 2001 and cultivated from 2002 on the site of INRA Clermont-Ferrand Theix (altitude of 870 m on a granitic brown soil) in monocultures in a fully factorial design with three replicate blocks, six cuts per year (6 cm above ground level) and two levels of nitrogen supply ( $N^- = 120$  and  $N^+ = 360$  kg N ha<sup>-1</sup> year<sup>-1</sup> in split applications at the beginning of each regrowth period) (Pontes *et al.*, 2007). For this study samples of the three replicates were harvested at the first cut in May 2005, and then dried in a forced air oven at 60 °C for 72h, before being ground through a 1 mm sieve.

Crude protein (CP) (Kjeldahl method), neutral-detergent fibre (NDF), acid-detergent fibre (ADF) and acid-detergent lignin (ADL) were determined for all samples. The *in-vivo* organic matter digestibility (OMD) was estimated by a pepsin-cellulase method (Aufrère *et al.*, 2007). *In-situ* degradation kinetics of the grass samples were performed on 3 rumen cannulated cows fed a diet of 70% hay and 30% concentrate. DegDM and DegN were calculated after fitting the *in situ* DM and N disappearance curves to the model of Ørskov and Mc Donald (1979) and assuming a particulate outflow rate from the rumen of 0.06 h<sup>-1</sup>. Species and N fertilization effects on rumen degradability were analysed by analysis of variance using the GLM procedure of SAS. When the treatment effect was significant ( $P < 0.05$ ) the means were compared by the Duncan multiple-range test.

## Results

The lowest CP contents were observed for *A. elatius*, *L. perenne*, *P. trivialis*, *A. odoratum* and the highest for *E. repens* and *D. glomerata*. CP content was increased on average by 10% at the high N fertilization level. The cell-wall contents (NDF, ADF) were the lowest for *L. perenne* and *P. trivialis*, and the highest were for *D. glomerata* and *F. rubra*. The OMD estimated by pepsin-cellulase digestibility was the highest for *L. perenne*, *P. trivialis*, and *A. odoratum* and the lowest for *F. rubra*. The increase in N fertilization did not modify the cell-wall contents or the OMD.

Table 1. Chemical composition (CP, NDF, ADF, ADL) in g kg<sup>-1</sup>DM, organic matter digestibility (OMD), rumen DM and N degradability (DegDM and DegN) in g g<sup>-1</sup>, estimated for the seven species at two levels of N fertilization (N<sup>-</sup> = 120 and N<sup>+</sup> = 360 kg N ha<sup>-1</sup>year<sup>-1</sup>).

Species	Fertilization	CP	NDF	ADF	ADL	OMD	DegDM	DegDN
<i>Lolium perenne</i>	N-	171	489	218	22.7	0.843	0.777	0.824
	N+	194	421	206	20.3	0.843	0.784	0.834
<i>Poa trivialis</i>	N-	170	478	219	34.8	0.811	0.777	0.814
	N+	175	470	207	26.4	0.820	0.752	0.778
<i>Festuca rubra</i>	N-	184	570	274	34.1	0.723	0.629	0.782
	N+	205	544	257	32.0	0.735	0.655	0.794
<i>Dactylis glomerata</i>	N-	193	577	268	34.8	0.791	0.699	0.757
	N+	213	572	274	41.1	0.796	0.719	0.750
<i>Anthoxanthum odoratum</i>	N-	179	528	238	24.3	0.808	0.751	0.819
	N+	198	553	247	25.9	0.808	0.747	0.834
<i>Elymus repens</i>	N-	211	543	248	31.5	0.782	0.707	0.802
	N+	236	558	266	39.2	0.780	0.679	0.777
<i>Arrhenatherum elatius</i>	N-	166	523	254	26.8	0.786	0.703	0.806
	N+	180	530	265	30.3	0.783	0.692	0.790
SEM							0.0062	0.0091
Species							$P < 0.001$	$P < 0.001$
Fertilization							$P > 0.05$	$P > 0.05$
Species * fertilization							$P < 0.05$	$P < 0.05$

In accordance with the OMD results, DegDM was significantly higher for *L. perenne*, *P. trivialis* and *A. odoratum*, and significantly lower for *F. rubra*. The DegN range was 0.78-0.83 g g<sup>-1</sup> for all grass species except *D. glomerata*, which was characterized by significantly lower nitrogen degradability. No significant effect of the fertilization level was found for DegDM and DegN, but the interaction between fertilization level and species was significant.

## Discussion

In accordance with the results of Pontes *et al.* (2007), the digestibility values of these native grasses were high (mean value of  $0.79 \text{ g g}^{-1}$ ) and close to that of a *L. perenne* cultivar (Clerpin) grown under the same conditions. The CP, cell-wall contents and digestibility values of the native *L. perenne* and *D. glomerata* sampled at a vegetative stage were close to the values reported in feed tables from INRA for improved cultivars (Baumont *et al.*, 2007). Furthermore, the phenological stage has an effect on chemical composition and ruminal degradability: *F. rubra* reached an advanced stage of maturity faster than the other species and had a low DegDM and OMD. As expected, within these grass species, the DegDM was positively related to the pepsin-cellulase digestibility ( $R^2 = 0.91$ ) and to a lesser extent negatively related to the cell-wall contents ( $R^2 = 0.69$  with ADF and  $R^2 = 0.53$  with NDF). In accordance with the results of Rodrigues *et al.* (2007) for permanent grasslands, the level of N fertilization had no effect on cell-wall contents and on OMD.

In contrast to the results obtained by (Nozières *et al.*, 2006) there was no significant relationship between DegN and CP content. The grass species with the highest DegN are also those with the highest DegDM and OMD (*L. perenne*, *P. trivialis* and *A. odoratum*). The exception is *D. glomerata* characterized by a lower N degradation in the rumen than the other grass species. This lower DegN may result from the action of polyphenol oxidase on protein degradation as high levels of this enzyme were found in *D. glomerata* by Lee *et al.* (2006). Lower DegN is interesting both for the ruminant nutrition by allowing a better protein utilization, and for the environment by reducing nitrogen losses. Furthermore, *D. glomerata* could contribute to improving the nitrogen value of permanent grasslands, as the low DegN is not associated with a low N content.

## Conclusions

The nutritive value of native grasses is thought to be lower than the nutritive value of forages genetically selected to obtain high yields, and this may be because phenological stage is more advanced in several species. Our results indicate that some native grasses have a nutritive value comparable to that of improved forage crops. The lower N degradability found in native *Dactylis glomerata* may contribute to improved protein value of semi-natural grasslands, with potential benefits both for the ruminant animal and for the environment.

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# Effects of seed mixture and N fertilization on nitrate content of grass-legume swards

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## Abstract

A trial was set up at the Institute for Animal Husbandry, Belgrade-Zemun, as a two-factor, randomized block design and conducted on four occasions during 2003 and 2004. The first factor in the investigations was sward type: pure lucerne (*Medicago sativa* L.) and three mixtures of lucerne (mixture I, with cocksfoot (*Dactylis glomerata* L.), mixture II, with cocksfoot and tall fescue (*Festuca arundinacea* Schreb.) and mixture III with cocksfoot, tall fescue and sainfoin (*Onobrychis viciifolia* Scop.). The second factor was fertilization, with four different amounts of N (0, 70, 140 and 210 kg N ha<sup>-1</sup>). Fertilization was carried out in two terms during a single year. The content of NO<sub>3</sub><sup>-</sup> in herbage of grass-legume swards in both years was largely dependent on fertilization and the interaction of the two factors, whereas the sward type had no statistical significance. Herbage nitrate content increased with an increase in the amount of added nitrogen. Treatments with 210 kg N ha<sup>-1</sup> resulted in a considerably higher concentration of NO<sub>3</sub><sup>-</sup> compared with the other treatments.

Keywords: grass-legume mixtures, nitrate, herbage quality

## Introduction

Nitrogen is an important constituent of plant compounds and its increased availability leads to an increase in herbage mass and improved herbage quality. Amounts of N added by fertilization need to be in accordance with the plant needs. The effect of fertilization depends on plant species in the mixture, since not all species react equally to the application of N fertilizer (Suleyman, 2003). Fertilization can increase the quality of forage of grass-legume mixtures (Ocokoljić *et al.*, 1984), but an excessive application of N mineral fertilizers can have adverse effects on quality since it leads to the accumulation of NO<sub>3</sub><sup>-</sup> in plants which is potentially harmful for animal health (Stanton, 2001). The content of nitrates in plant material becomes dangerous when nitrogen doses become considerably higher than what is necessary to provide satisfactory yield of dry matter. Nitrate content varies from cut to cut and with the type of crop and fertilization (Shiel *et al.*, 1999). The objective of the research was to determine if N fertilization of lucerne (*Medicago sativa* L.) and its mixtures with cocksfoot (*Dactylis glomerata* L.) tall fescue (*Festuca arundinacea* Schreb.) and sainfoin (*Onobrychis viciifolia* Scop.) causes nitrate accumulation, which can have harmful effects on forage quality and on the health of the livestock consuming this feed.

## Materials and methods

A two-factor trial was set at the Institute for Animal Husbandry, Belgrade-Zemun during 2003-2004. The first factor in the investigations was sward type: pure lucerne and its three mixtures (mixture I, with cocksfoot; mixture II, with cocksfoot and tall fescue; and mixture III with cocksfoot, tall fescue and sainfoin. The second factor was fertilization with four different quantities of N (0, 70, 140 and 210 kg N ha<sup>-1</sup>). Fertilization was carried out on two occasions during a single year, in 2003 after sowing and after the first cut, and in 2004 at the beginning



of vegetation and after the first cut. The grassland was cut three times in the first year, and four times in the second. The nitrate content in the herbage DM of mixtures was investigated as a quality parameter and is presented in this paper as an average value of all cuts in the investigation year. The data were analysed by analysis of variance for a 4 x 4 randomized block design with four blocks. When  $P < 0.05$ , pairwise comparisons among the treatment means were performed using the LSD test.

The soil on which the trial was set was poor carbonate chernozem, of favourable water, air and thermal regime, and very good granular structure. Chemical characteristics of the soil are presented in Table 1.

Table 1. Chemical characteristics of the soil on which the trial was conducted.

Depth cm	pH H <sub>2</sub> O	pH KCl	Humus g kg <sup>-1</sup>	CaCO <sub>3</sub> g kg <sup>-1</sup>	N Total mg kg <sup>-1</sup>	N-NO <sub>3</sub> mg kg <sup>-1</sup>	P <sub>2</sub> O <sub>5</sub> mg kg <sup>-1</sup>	K <sub>2</sub> O mg kg <sup>-1</sup>
0-20	7.29	7.08	43.5	3.3	1975	52	909	162
20-40	7.23	7.09	44.3	4.7	1938	57	918	165

Average air temperatures during 2003 and 2004 were higher than the long-term average and were recorded as 12.7 °C and 12.4 °C, respectively. Based on the data on total annual precipitation it can be concluded that 2003 was dry with 551.9 mm of precipitation, which was 93.3 mm lower than the average value determined for several years. In 2004, however, the precipitation was as high as 831.6 mm which had a positive effect on the productive characteristics of plants.

## Results and discussion

The content of NO<sub>3</sub><sup>-</sup> in grass-legume swards in both years was largely dependent on the fertilization. The first year was characterized by a higher nitrate content in plants, 21.3% more, or 459.1 mg kg<sup>-1</sup>, compared with the second year (Table 2). This can be explained by the dry weather conditions leading to the accumulation of NO<sub>3</sub><sup>-</sup> (Stanton, 2001). This agrees with the results obtained by Totev *et al.* (1997).

The sward type had no statistically significant effect on nitrate content in plant material in either of the investigation years.

Treatments with 210 kg N ha<sup>-1</sup> resulted in considerably higher concentrations of NO<sub>3</sub><sup>-</sup> compared with other treatments, as well as treatments with 140 kg N ha<sup>-1</sup> and 70 kg N ha<sup>-1</sup> compared with the treatment without N. Jeremić and Stošić (1981) also concluded in their research that the content of NO<sub>3</sub><sup>-</sup> in mixture increased with the addition of N, i.e. with 30 kg N ha<sup>-1</sup>, the content of NO<sub>3</sub>-N was 19 mg kg<sup>-1</sup>, with addition of 180 kg N ha<sup>-1</sup>, the content of NO<sub>3</sub>-N increased to 30 mg kg<sup>-1</sup>. The 2003 treatment with 210 kg N ha<sup>-1</sup> resulted in the greatest accumulation of NO<sub>3</sub><sup>-</sup> in the herbage, with 186% more NO<sub>3</sub><sup>-</sup> than the treatment without N. Treatments without N had the lowest content of NO<sub>3</sub><sup>-</sup>. In the second year, treatments with the highest amount of N resulted in 289% more NO<sub>3</sub><sup>-</sup> than the treatment without N. The interaction of factors in both investigation years had a significant effect on the content of NO<sub>3</sub><sup>-</sup> in swards. Differences in content of NO<sub>3</sub><sup>-</sup> occurred mainly between treatments without N and treatments with 210 N kg ha<sup>-1</sup> within certain sward types. The highest content in the first year was established in mixture II with 210 kg N ha<sup>-1</sup> of 3,812 mg kg<sup>-1</sup>, and in the second investigation year in mixture III fertilized with 210 kg N ha<sup>-1</sup> of 3,479 mg kg<sup>-1</sup>. Higher accumulation of NO<sub>3</sub><sup>-</sup> in swards fertilized with N compared to pure lucerne is explained by physiological predestination of grasses to accumulate higher NO<sub>3</sub><sup>-</sup> concentration, whereas differences between swards occur as a consequence of higher or lower incidence of weed plants capable of accumulation of sufficient amount of NO<sub>3</sub><sup>-</sup> (Stanton, 2001).



Table 2. Average content of  $\text{NO}_3^-$  ( $\text{mg kg}^{-1}$ ) in DM of sown grassland depending on the sward type and N fertilization over cuts in 2003 and 2004.

Year	N fertilization ( $\text{kg ha}^{-1}$ )(B)	Sward type (A)				Average
		Lucerne	Mixture I	Mixture II	Mixture III	
2003	0	1054	1175	1317	1215	1190
	70	1694	1350	1939	1820	1701
	140	1895	2808	2366	2227	2324
	210	3055	3227	3812	3508	3401
	Average	1925	2140	2358	2192	
2004	0	836	593	652	846	732
	70	1271	1441	839	1300	1213
	140	1875	2167	1698	2215	1989
	210	2550	2972	2386	3479	2847
	Average	1633	1793	1394	1960	
		B		AB		
2003	$\text{LSD}_{0.05}$	654*		1774*		
2004	$\text{LSD}_{0.05}$	704*		1469*		

\*significant at  $P < 0.05$ ; A-effect of sward type; B-effect of N fertilization; AB-interaction between sward type and N fertilization.

The UK Agricultural Research Council (1980) (cited in Shiel *et al.*, 1999) stated that concentrations higher than 3000-5000  $\text{mg kg}^{-1}$  of  $\text{NO}_3^-$  can be regarded as potentially dangerous and should be avoided in certain types/groups of animals, e.g. pregnant livestock because of the risk of miscarriage. In the research presented here certain treatments resulted in concentrations which exceeded this limit. Thus, it can be concluded that fertilization with high amounts of N results in feed of poor quality which is harmful for certain types of livestock

## Conclusions

The content of  $\text{NO}_3^-$  in lucerne and grass-legume mixtures in both investigation years was largely dependent on the fertilization and the interaction of the investigated factors. With an increase in the amount of N the content of  $\text{NO}_3^-$  increased. The highest  $\text{NO}_3^-$  content in both investigation years was established in treatments receiving 210  $\text{kg N ha}^{-1}$ . These treatments caused an increase of  $\text{NO}_3^-$  content, in the first year by 186%, and in the second year by 289%. With a view to producing higher yields, large quantities of N are often used. However, such quantities can cause accumulation of  $\text{NO}_3^-$  in forage which is potentially harmful for livestock health. Therefore, it is necessary to reduce N fertilization to the detriment of yield but in favour of quality.

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# Contents of $\alpha$ -tocopherol and $\beta$ -carotene in grasses and legumes harvested at different maturities

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## Abstract

Concentrations of  $\alpha$ -tocopherol and  $\beta$ -carotene in forage species at various maturities were studied in Scandinavia. Red clover (RC)/timothy (TI), RC/meadow fescue (MF), and birdsfoot trefoil (BT)/TI mixtures were grown in Skara and Umeå, Sweden. RC/TI, RC/perennial ryegrass (PR), white clover/PR and BT/TI were grown in Foulum, Denmark. Forages in Sweden were cut one week before heading (BH), at heading and one week after heading of TI. The regrowth was cut six and eight weeks after each harvest in the spring growth cycle. In Denmark, one first harvest and three regrowths were taken. Results from Skara and Foulum are presented. Highest concentrations of  $\alpha$ -tocopherol and  $\beta$ -carotene (mg kg<sup>-1</sup> DM) in legumes were found in BT grown in Skara (49.8 and 69.6 in spring growth cycle, 48.1 and 79.8 in regrowth) and in Foulum (81.3 and 89.2). MF had more  $\alpha$ -tocopherol and  $\beta$ -carotene than TI in the spring growth cycle (73.5 and 54.2 vs. 46.9 and 43.0 mg kg<sup>-1</sup> DM). Highest concentrations of vitamins in the regrowth were found six weeks after BH with 71.8 and 104.8 mg  $\alpha$ -tocopherol and 99.6 and 73.1 mg  $\beta$ -carotene kg<sup>-1</sup> DM in legumes and grasses, respectively.

Keywords: forages, vitamins, maturity stage, season

## Introduction

Forage is the largest natural source of vitamins for ruminants (NRC, 2001), but the knowledge about how forage vitamin content is affected by species, maturity stage and climate is limited. Shortage of vitamins A and E can depress the immune system and cause reproductive disturbances in ruminants (NRC, 2001). The objective of this experiment was to determine the effects of plant species, maturity stage, season, and the interactions between plant species and maturity stage and plant species and season, on concentrations of  $\alpha$ -tocopherol and  $\beta$ -carotene in forages grown in Sweden and Denmark.

## Materials and methods

Plots with mixtures of grasses and legumes were established in Skara and Umeå, Sweden and in Foulum, Denmark in the spring of 2004. This paper includes results from Skara and Foulum. Mixtures grown in Sweden were: 1) timothy (TI; *Phleum pratense* L.)/birdsfoot trefoil (BT; *Lotus corniculatus* L.), 2) TI/red clover (RC; *Trifolium pratense* L.), and 3) meadow fescue (MF; *Festuca pratensis* L.)/RC. Harvest dates during the spring growth cycle in Skara 2005 were one week before (1:1; June 9) at (1:2; June 15) and one week after (1:3; June 22) heading of timothy. Regrowths were cut six and eight weeks after 1:1 (July 20 and

August 3), 1:2 (July 26 and August 8) and 1:3 (August 2 and August 17). Mixtures grown in Foulum were TI/RC, TI/BT, perennial ryegrass (PR; *Lolium perenne* L.)/RC and PR/white clover (WC; *Trifolium repens* L.). Forages were harvested May 30 and regrowths were harvested July 5, August 22 and October 24 at Foulum. Dry-matter (DM) yields were determined at both locations. All mixtures were separated into species before analysis. Alpha-tocopherol and  $\beta$ -carotene in grasses and legumes were analysed at Foulum, Denmark by HPLC, after saponification and extraction into heptane (Jensen *et al.*, 1998). The trial in Skara had a split-plot design with mixture treated as the main plot, replicated three times, and with spring growth cycle and regrowth as sub plots. The trial in Foulum had a randomized block design with mixture, replicated four times, and harvest number as fixed factors. Data were analysed for grasses and legumes separately using the general linear model procedure of SAS (ver. 9.1, 2001).

## Results and discussion

In Skara, interactions were detected between legume species and cutting time during the spring growth cycle ( $P = 0.001$ ), where the  $\alpha$ -tocopherol content decreased in BT but increased in RC, grown with TI, with later harvest date. As a result, BT had higher levels of  $\alpha$ -tocopherol in mg kg<sup>-1</sup> DM than RC at 1:1 (63.9 vs. 19.9) and 1:2 (49.6 vs. 24.1) but there was no difference between the legumes at 1:3 (35.9 vs. 31.5). Concentrations of  $\beta$ -carotene in legumes were highest at 1:2, whereas 1:2 only differed from 1:3 in grasses (Table 1). MF had higher levels of  $\alpha$ -tocopherol and  $\beta$ -carotene than TI, when averaged over harvests, which can be related to a larger proportion of leaf in the DM in MF than in TI (60 vs. 30% of DM) as vitamins are found primarily in the leaves of plants.

Table 1. Alpha-tocopherol and  $\beta$ -carotene, mg kg<sup>-1</sup> DM, in the spring growth cycle of grasses and legumes grown in Skara.

	Mixture			<i>P</i> -value	Harvest time			<i>P</i> -value
	1	2	3		1:1	1:2	1:3	
<i>Grasses</i>	<i>TI</i>	<i>TI</i>	<i>MF</i>					
$\alpha$ -tocopherol	50.6 <sup>b</sup>	43.1 <sup>b</sup>	73.5 <sup>a</sup>	0.001	61.0	55.6	50.6	ns
$\beta$ -carotene	43.0 <sup>b</sup>	42.9 <sup>b</sup>	54.2 <sup>a</sup>	0.028	48.0 <sup>a</sup>	53.4 <sup>a</sup>	38.6 <sup>b</sup>	0.003
<i>Legumes</i>	<i>BT</i>	<i>RC</i>	<i>RC</i>					
$\alpha$ -tocopherol	49.8 <sup>a</sup>	24.7 <sup>b</sup>	24.4 <sup>b</sup>	0.0004	34.6	32.6	31.8	ns
$\beta$ -carotene	69.6 <sup>a</sup>	48.5 <sup>b</sup>	42.9 <sup>b</sup>	0.005	48.8 <sup>b</sup>	68.3 <sup>a</sup>	43.9 <sup>b</sup>	0.002

TI = timothy, MF = meadow fescue, BT = birdsfoot trefoil, RC = red clover.

1:1 = one week before heading, 1:2 = at heading, 1:3 = one week after heading of timothy in spring growth cycle

<sup>a, b</sup>Means with different superscripts within the same row differ significantly ( $P < 0.05$ ).

In the regrowth of grasses and legumes, as a mean over species, the highest levels of both  $\alpha$ -tocopherol and  $\beta$ -carotene were found six weeks after 1:1 (Table 2).

Table 2. Alpha-tocopherol and  $\beta$ -carotene, mg<sup>-1</sup> kg DM, in the regrowth of grass and legume harvested in Skara six and eight weeks after (w a) first cut.

	Regrowth						<i>P</i> -value
	6 w a 1:1	8 w a 1:1	6 w a 1:2	8 w a 1:2	6 w a 1:3	8 w a 1:3	
<i><math>\alpha</math>-tocopherol</i>							
Grass	104.8 <sup>a</sup>	73.7 <sup>b</sup>	68.6 <sup>b</sup>	41.4 <sup>c</sup>	54.0 <sup>c</sup>	54.5 <sup>c</sup>	0.0036
Legume	71.8 <sup>a</sup>	47.5 <sup>b</sup>	37.7 <sup>c</sup>	23.0 <sup>d</sup>	25.0 <sup>d</sup>	38.7 <sup>c</sup>	< 0.0001
<i><math>\beta</math>-carotene</i>							
Grass	73.1 <sup>a</sup>	46.7 <sup>c</sup>	34.2 <sup>d</sup>	32.9 <sup>d</sup>	38.7 <sup>c</sup>	60.4 <sup>b</sup>	< 0.0001
Legume	99.6 <sup>a</sup>	63.8 <sup>c</sup>	56.0 <sup>c</sup>	43.3 <sup>d</sup>	26.5 <sup>e</sup>	88.0 <sup>b</sup>	< 0.0001

1:1 = one week before heading, 1:2 = at heading, 1:3 = one week after heading of timothy in spring growth cycle

<sup>a, b, c, d</sup>Means with different superscripts within the same row differ significantly ( $P < 0.05$ ).

On average, MF had a higher level of  $\alpha$ -tocopherol in  $\text{mg kg}^{-1}$  DM than TI in the regrowth after the early cut (1:1) in first harvest (121.2 vs. 74.2) and after the late cut (1:3) in the first harvest (63.5 vs. 49.6;  $P = 0.002$ ). In the average regrowth after 1:2, MF only differed from TI grown with BT (62.8 vs. 47.3  $\text{g kg}^{-1}$  DM). Averaged over 1:1, 1:2 and 1:3, MF had a higher level of  $\alpha$ -tocopherol in  $\text{mg kg}^{-1}$  DM than TI at six (98.6 vs. 64.4) and eight weeks (66.3 vs. 51.6) of regrowth ( $P = 0.02$ ). In legumes, BT had a higher level of  $\alpha$ -tocopherol than RC six weeks after first harvest (51.7 vs. 38.8  $\text{mg kg}^{-1}$  DM), but there was no difference between species eight weeks after first harvest. Average total herbage yield of TI/BT was 8,583 kg DM, compared with 10,133 kg DM  $\text{ha}^{-1}$  of TI/RC.

In Denmark, interactions between harvest and species were found in concentrations of  $\alpha$ -tocopherol and  $\beta$ -carotene of legumes and in the  $\beta$ -carotene concentration of grasses ( $P < 0.001$ ). BT had a higher level of  $\alpha$ -tocopherol in  $\text{mg kg}^{-1}$  DM than the other legumes at the second (74.1 vs. 34.8), third (70.6 vs. 20.2) and fourth harvest (118.4 vs. 64.2). High concentrations of vitamins in herbage at the fourth harvest indicate that climate, in addition to phenological composition of plants, plays an important role in vitamin content of forage (Table 3). TI contained more  $\alpha$ -tocopherol in  $\text{mg kg}^{-1}$  DM than PR (78.4 vs. 53.3) and BT contained more  $\alpha$ -tocopherol than white and red clover (81.3 vs. 41.6;  $P < 0.0001$ ), when averaged over harvests. The content of  $\beta$ -carotene in BT, WC and RC was 89.2, 74.4 and 52.3  $\text{mg kg}^{-1}$  DM, respectively ( $P < 0.0001$ ). Average total yield of TI/BT was 7,025 kg DM compared with 11,576 kg DM  $\text{ha}^{-1}$  of TI/RC.

Table 3. Alpha-tocopherol and  $\beta$ -carotene,  $\text{mg kg}^{-1}$  DM, in grass and legume harvested in Foulum, Denmark on May 30 (1), July 5 (2), August 22 (3) and October 24 (4) 2005.

	Harvest				P-value
	1	2	3	4	
<i><math>\alpha</math>-tocopherol</i>					
Grass	54.4 <sup>b</sup>	41.5 <sup>c</sup>	38.0 <sup>c</sup>	104.3 <sup>a</sup>	< 0.0001
Legume	50.8 <sup>b</sup>	44.6 <sup>bc</sup>	32.8 <sup>c</sup>	77.7 <sup>a</sup>	< 0.0001
<i><math>\beta</math>-carotene</i>					
Grass	30.5 <sup>b</sup>	30.0 <sup>b</sup>	39.7 <sup>b</sup>	152.2 <sup>a</sup>	< 0.0001
Legume	40.7 <sup>b</sup>	46.2 <sup>b</sup>	39.2 <sup>b</sup>	142.0 <sup>a</sup>	< 0.0001

<sup>a,b,c</sup>Means with different superscripts within the same row differ significantly ( $P < 0.05$ )

## Conclusions

BT had a higher concentration of vitamins than RC and WC, but the yield was lower than in RC. Highest vitamin levels in the regrowth herbage were achieved when the cut was taken six weeks after the first early cut (1:1). Higher levels of vitamins in herbage harvested in the autumn, compared with the other harvest dates, indicate that vitamins are affected by climate.

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# Effect of plant mediated proteolysis on protein degradation among ten diploid perennial ryegrass (*Lolium perenne* L.) genotypes

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## Abstract

Forage feeding systems for ruminants are often characterized by low N-use efficiency resulting in high N-loss to the environment, especially under grazing conditions. Protein degradation in the rumen is primarily ascribed to microbial activity, but recent investigations revealed that plant proteases may play a role as well. The proteolytic activity in plants varies among different forage species. The discovered genetic variations between genotypes of perennial ryegrass (PR) may set a new basis for future breeding targets concerning forage quality in order to support higher N-use efficiency in the rumen.

The objective of the present study is to quantify plant-mediated protein degradation of ten diploid PR genotypes from the early heading date in the first and second cut in two subsequent years. The experiment was conducted (randomized complete block design with three replicates) in Northern Germany during 2006 and 2007. Leaf blades were sampled in the field, sterilized and incubated under anaerobic conditions at 39 °C for 0, 6 and 24 h. Protein content in leaves was determined by Bradford analysis.

Results revealed that the protein concentration declined significantly with increasing the incubation period. However, the calculated degradation rate in both years did not significantly differ between the genotypes.

Keywords: proteolysis, protease, fresh forage

## Introduction

Perennial ryegrass (PR) belongs to the most important grass family in the northwest European region. Although it is characterized by a high yielding potential accompanied by high energy content, grassland based production systems are known to be low in N utilization of ruminants, leading to a major loss of N to the environment. It is assumed that protein degradation in rumen is carried out entirely by microbes in the rumen. Recent investigations revealed that the contribution of plant-mediated proteolytic enzymes contained in ingested forage have been ignored in current models of protein degradation (Theodorou *et al.*, 1996; Kingston Smith *et al.*, 2003). The majority of ingested plant biomass of the grazing ruminant consists of intact cells, which stay viable and metabolically active during the first hours after ingestion. These cells contain proteases in their vacuoles which are involved in proteolysis. Protease activity can be induced by damage or biotic stress. Conditions found in the rumen (elevated temperature, darkness, and lack of O<sub>2</sub>) may contribute to induce the plant cell death and increase plant-mediated protease activity (Attwood, 2005). Theodorou *et al.* (1996) and Kingston-Smith and Theodorou (2000) stated that the intact cells entering the rumen could undergo an enforced, premature senescence. The plant protease activity varies between different plant species (Pichard *et al.*, 2006). However, there is lack of data about differences between the protein degradability and availability among perennial ryegrass genotypes. In this study the response of ten PR genotypes to similar rumen conditions is tested by using an *in-vitro* system, which simulates the main stresses of the rumen but from which rumen microbes



are excluded. The objective was to identify genotypes which are characterized by a higher protein quality as a result of decreased plant-mediated protease activity.

## Materials and methods

The samples for this analysis were collected from a field trial in Northern Germany during the growing seasons 2006 and 2007. Ten different diploid PR genotypes belonging to the intermediate-early heading group were grown in a randomized complete block experimental design with three replicates. Nitrogen fertilizer was applied at the rate of 300 kg ha<sup>-1</sup> y<sup>-1</sup>. Leaf blades of the first and second cuts were harvested in June/July 2006 and May/June 2007. Residual soil particles were removed by washing the sampled plant material several times in distilled water. The material was also sterilized in 80% ethanol to avoid microbial growth during incubation. Ruminant mastication was simulated by cutting the leaves into approximately 2 cm length. The foliage of each genotype were separated into three parts each consisting of 1g fresh matter (FM) in three replications. The first part was immediately frozen in liquid nitrogen and then stored at -70 °C without incubation until required. The two other parts were incubated in darkness under anaerobic conditions at 39 °C for 6 h and 24 h, respectively, in bottles filled with 100 ml incubation buffer (50 mM Na<sub>2</sub>HPO<sub>4</sub>, 50 mM KH<sub>2</sub>PO<sub>4</sub>, 5 mM dithiothreitol [DTT], 2% sodium azide, pH 6.8). At the end of each incubation period foliage residues were separated from incubation buffer by filtration through a filter and immediately frozen in liquid nitrogen and stored at -70 °C until required. The residual protein content of the samples was measured according to the method described by Kingston Smith (2003). The frozen leaflets were ground to a fine powder in liquid nitrogen with pestle and mortar. Sub-samples of 0.3 to 0.5 g were weighed into micro tubes and 800 µl extraction buffer (50 mM Tris, 2 mM DTT, 1mM EDTA, 0.1% Triton X 100, pH 7.5) was added. These sub-samples were homogenized and centrifuged for 10 min at 10,000 g. The protein content of the supernatant was determined according to Bradford (1976). The degradation rate was calculated with the following equation: [Kd (%/h) = (ln B<sub>0</sub> – ln B<sub>24</sub>)/24], where B<sub>0</sub> and B<sub>24</sub> are the protein contents after 0 hours and 24 hours. The influence of the single factors genotype (G), incubation period (IP) and cut (C) on the protein content (µg g<sup>-1</sup> FM) were submitted to analysis of variance. Means were separated by LSD at *P* < 0.05 and probabilities corrected with Bonferroni-Holm Test.

## Results and discussion

Analysis of variance revealed that the interactions between the three studied factors influenced the protein content in 2006. Protein content of genotypes varied from 37.3 µg g<sup>-1</sup> FM to 58.7 µg g<sup>-1</sup> FM. However, in 2007 no significant variations were observed among the tested genotypes. The changes in the average protein content during incubation are presented

Table 1. Changes in the average protein content (µg g<sup>-1</sup> FM) during incubation.

Hours	Cutting dates			
	1 <sup>st</sup> cut 2006	2 <sup>nd</sup> cut 2006	1 <sup>st</sup> cut 2007	2 <sup>nd</sup> cut 2007
0	72.1 <sup>a</sup>	126.7 <sup>a</sup>	115.7 <sup>a</sup>	67.6 <sup>a</sup>
6	24.8 <sup>b</sup>	35.7 <sup>b</sup>	37.6 <sup>b</sup>	18.0 <sup>b</sup>
24	7.9 <sup>c</sup>	14.7 <sup>c</sup>	14.2 <sup>c</sup>	4.5 <sup>c</sup>
SE = 3.3		SE = 2.5		

Means followed by the same letter within the same column are not significantly different at *P* < 0.05.

in Table 1. The protein content showed significantly superior values when no incubation was done (0h) for the two cutting dates. The tested variable significantly declined with increasing incubation period, and was lowest when incubated for 24 hours under the two cutting dates. The amount of protein losses from the tissues over 24 h accounts

to 89% and 88% in the year 2006, and 88% and 93% in the year 2007, for the first and second cuts respectively. These results support the findings of Beha *et al.* (2002) who reported a protein decline of 82.3% after 24 hours incubation under the same conditions for perennial ryegrass.

The calculated degradation rates of the ten tested genotypes are presented in Table 2. In both years there were no observed significant differences between the tested genotypes. The values fluctuate in the range of 6.9 to 12.4  $\mu\text{g g}^{-1}$  FM per hour in the year 2006 and from 6.7 to 12.2  $\mu\text{g g}^{-1}$  FM per hour in the year 2007. The measured protein contents exceeded findings of Beha *et al.* (2002) who stated that one-third of the initial protein is degraded after three hours incubation.

Table 2. Decline in protein content ( $\mu\text{g g}^{-1}$  FM) per hour of the ten genotypes.

Genotype	1 <sup>st</sup> cut 2006	2 <sup>nd</sup> cut 2006	1 <sup>st</sup> cut 2007	2 <sup>nd</sup> cut 2007
1	12.4 <sup>a</sup>	8.5 <sup>a</sup>	6.7 <sup>a</sup>	12.2 <sup>a</sup>
2	9.5 <sup>a</sup>	8.1 <sup>a</sup>	8.8 <sup>a</sup>	9.0 <sup>a</sup>
3	10.3 <sup>a</sup>	6.9 <sup>a</sup>	8.2 <sup>a</sup>	10.6 <sup>a</sup>
4	9.5 <sup>a</sup>	7.5 <sup>a</sup>	7.5 <sup>a</sup>	10.3 <sup>a</sup>
5	11.1 <sup>a</sup>	8.1 <sup>a</sup>	9.9 <sup>a</sup>	12.0 <sup>a</sup>
6	11.5 <sup>a</sup>	8.6 <sup>a</sup>	9.7 <sup>a</sup>	10.4 <sup>a</sup>
7	8.1 <sup>a</sup>	8.2 <sup>a</sup>	7.1 <sup>a</sup>	9.0 <sup>a</sup>
8	9.0 <sup>a</sup>	10.2 <sup>a</sup>	7.1 <sup>a</sup>	11.1 <sup>a</sup>
9	7.9 <sup>a</sup>	9.5 <sup>a</sup>	8.9 <sup>a</sup>	12.1 <sup>a</sup>
10	8.9 <sup>a</sup>	7.0 <sup>a</sup>	8.0 <sup>a</sup>	10.6 <sup>a</sup>
	SE = 1.6		SE = 1.0	

Means followed by the same letter within the same column are not significantly different at  $P < 0.05$ .

Main effect for cutting dates were significant for each year ( $P < 0.05$ ).

## Conclusion

The significant decline in protein content along the incubation period confirmed that the plant-mediated proteases are involved in the process of proteolysis. However, genotypes with similar ploidy and heading date showed similar degradation rates.

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# Agronomic potential of tanniferous forage plant species and cultivars

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## Abstract

Condensed tannins (CT) can have beneficial effects on ruminant health and productivity and can contribute to a reduction in dietary methane and ammonia emissions. Thus the agronomic potential of tanniferous (tanniniferous) forage plant species and their cultivars are of interest. In our two-year field experiment *Onobrychis viciifolia* Scop. (sainfoin) was the most promising species with high tannin concentrations of up to 102 g CT kg<sup>-1</sup> dry matter and large forage yields of up to 9.9 Mg ha<sup>-1</sup> y<sup>-1</sup> dry matter when grown in monoculture. Due to lower CT concentrations, *Lotus corniculatus* L. (birdsfoot trefoil) revealed a lower potential as a tanniferous forage species. Unsatisfactory potentials were found for *Cichorium intybus* L. (chicory) due to very low tannin concentrations and for *Lotus pedunculatus* Cav. (big trefoil) because of very small yield. The low competitive ability of tanniferous plant species reduced their forage yield considerably when grown in mixture with *Festuca pratensis* Hudson (meadow fescue). Among the three cultivars tested from each of the tanniferous species, important variabilities in tannin concentration and competitive ability were detected, suggesting that the agronomic potential of these species can be improved by plant breeding.

Keywords: condensed tannins, genetic variability, yield, mixture, competitive ability

## Introduction

Condensed tannins (CT) in forage can have beneficial effects on the productivity and health of ruminants (Barry and McNabb, 1999; Min *et al.*, 2003). They can improve protein supply, reduce the risk of bloat, decrease the burden of gastrointestinal nematodes and reduce the emissions of methane and ammonia. The effect against parasites was observed in fresh material, hay and silage (Heckendorn *et al.*, 2006).

Tanniferous plant species and cropping techniques that enable a large and stable forage yield with a satisfactory concentration of CT are the prerequisites for a successful use of forage containing CT. Thus the aim of our two-year field experiment was to test the yielding capacity and the tannin concentration of twelve cultivars of four tanniferous (tanniniferous) forage plant species grown as monocultures and in simple mixtures.

## Materials and methods

The experiment was conducted at the Agroscope Reckenholz-Tänikon Research Station ART, Zurich, Switzerland (47°26'N, 8°30'E; elevation 440 m). Monocultures with three varieties of *Onobrychis viciifolia* Scop. (sainfoin), *Lotus corniculatus* L. (birdsfoot trefoil), *Lotus pedunculatus* Cav. (big trefoil), and *Cichorium intybus* L. (chicory) were sown. The seed densities of 180, 18, 18 and 4 kg ha<sup>-1</sup>, respectively, were adjusted for germination percentage. For the mixtures, 40% of the seed of the respective tanniferous species was replaced by 10 kg ha<sup>-1</sup> of seed of *Festuca pratensis* Hudson (meadow fescue) cv. 'Préval'. Plots containing *C. intybus* received 50 kg ha<sup>-1</sup> mineral N fertilizer for each cut.

Table 1. Total yield, yield of tanniferous plants and their CT concentration: 12 cultivars of 4 different species, in monoculture and in mixture with *Festuca pratensis*. Mean  $\pm$  se; n = 3.

	Total herbage Yield (Mg ha <sup>-1</sup> y <sup>-1</sup> DM)		Tanniferous plant Yield (Mg ha <sup>-1</sup> y <sup>-1</sup> DM)		CT concentration (g CT kg <sup>-1</sup> DM)
	Pure	mixture	pure	mixture	
<i>O. viciifolia</i>					
Alvaschein	11.0 ± 0.3	16.4 ± 1.1	5.1 ± 1.0	1.1 ± 0.3	102 ± 26
Commercial seed	11.1 ± 0.2	16.4 ± 1.1	4.3 ± 1.5	1.0 ± 0.5	83 ± 17
Visnovsky	13.0 ± 1.2	16.5 ± 1.6	9.9 ± 1.3	6.0 ± 1.0	67 ± 17
<i>L. pedunculatus</i>					
Maku	7.7 ± 0.3	18.1 ± 1.0	1.5 ± 0.3	1.0 ± 0.4	46 ± 16
Barsilvi	9.0 ± 1.1	17.1 ± 0.1	0.2 ± 0.1	0.2 ± 0.1	44 ± 14
Sunrise	8.6 ± 1.1	17.6 ± 1.0	0.4 ± 0.1	0.1 ± 0.1	30 ± 10
<i>L. corniculatus</i>					
Odenwlder	9.9 ± 1.0	18.0 ± 1.2	7.0 ± 1.8	3.7 ± 1.4	25 ± 7
Lotar	11.0 ± 1.0	18.4 ± 1.1	8.5 ± 1.0	5.8 ± 1.7	14 ± 6
Oberhaunstdter	10.0 ± 1.5	17.3 ± 1.1	7.6 ± 1.0	6.5 ± 1.5	9 ± 1
<i>C. intybus</i>					
Puna	12.8 ± 1.0	12.8 ± 0.6	12.8 ± 1.0	12.0 ± 0.6	6 ± 1
Forage Feast	9.9 ± 0.5	12.6 ± 0.7	9.8 ± 0.5	9.2 ± 1.5	6 ± 1
Lacerta	12.1 ± 1.2	12.5 ± 1.5	12.0 ± 1.2	10.9 ± 1.4	4 ± 1

Plots containing legumes received 25 kg ha<sup>-1</sup> N per cut. In the sowing year 2004, the establishing swards were cut twice. In 2005, dry matter (DM) yield was determined for each of the sown species and for unsown species: the plots were harvested four times with a 'Hege 212' plot-harvester at 7 cm cutting height. In spring 2006, pure herbage of tanniferous species was sampled for analysis of the CT concentration as described in Hring *et al.* (2007b). The effects of species, variety and mixture were tested by analysis of variance.

## Results and discussion

The performances in yield and CT concentration differed widely among the assessed plant species ( $P < 0.01$ , Table 1). In monoculture the slowly developing *L. pedunculatus* was out-competed by unsown species and in mixture by *F. pratensis*, resulting in very small yields of 0.2 to 1.5 Mg ha<sup>-1</sup> y<sup>-1</sup> and 0.2 to 1.0 Mg ha<sup>-1</sup> y<sup>-1</sup>, respectively. Although *C. intybus* was large-yielding, it had a very low CT concentration (4 to 6 g CT kg<sup>-1</sup> DM). On the basis of these results and the experimental conditions, *L. pedunculatus* and *C. intybus* were considered to have an insufficient potential as tanniferous forage plants.

The other two species examined showed reasonable to good potential for use as tanniferous forage plants. Grown in monoculture *L. corniculatus* performed well (7.0 to 8.5 Mg ha<sup>-1</sup> y<sup>-1</sup>, Table 1) and suppressed the invasion of unsown species successfully showing a high yield proportion of 60 to 90% of the total forage harvested (Fig. 1). In mixture, *L. corniculatus* yielded 3.7 to 6.5 Mg ha<sup>-1</sup> y<sup>-1</sup>. The cultivar 'Visnovsky' of *O. viciifolia* showed a large yield production with 9.9 Mg ha<sup>-1</sup> y<sup>-1</sup> in monoculture and 6.0 Mg ha<sup>-1</sup> y<sup>-1</sup> in mixture. The concentration of condensed tannins in the plant material was high (67 to 102 g CT kg<sup>-1</sup> DM) in *O. viciifolia* and reasonable in *L. corniculatus* (9 to 25 g CT kg<sup>-1</sup> DM). This is in agreement with Hring *et al.* (2007b).

The mixtures with *F. pratensis* resulted in larger total yields than the monoculture (Table 1) with a clear suppression of unsown species (Hring *et al.*, 2007a), but *F. pratensis* was quite competitive in the mixtures. Consequently, in mixture, absolute yields ( $P < 0.0001$ , Table 1) and yield proportions ( $P < 0.0001$ , Figure 1) of the tanniferous species were clearly lower than in monoculture. The competitive ability of *F. pratensis* was especially strong during the reproductive growth until June, as reflected in the much lower proportion of tanniferous plants in the mixture than in the monoculture (Figure 1). Focusing on production of

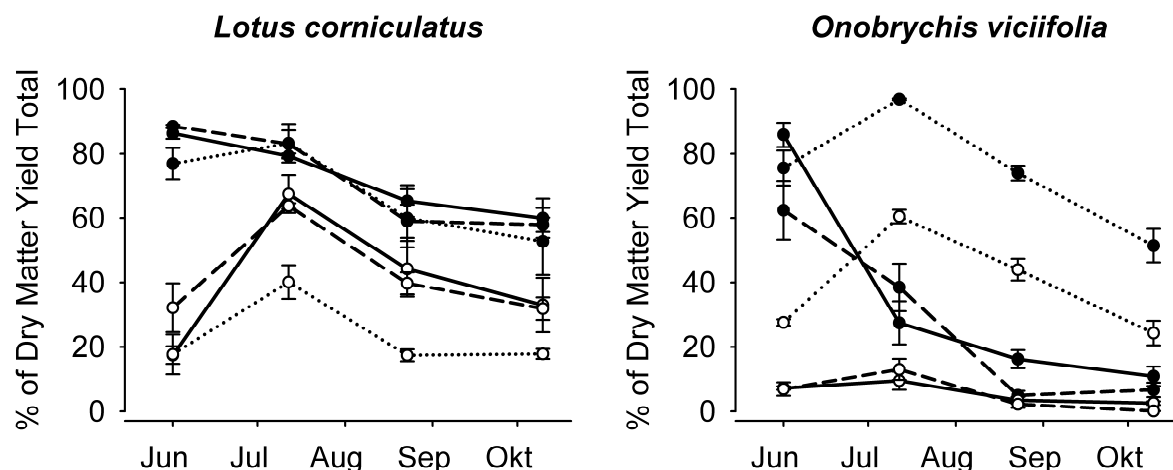


Figure 1. Yield proportion of *L. corniculatus* cv. Lotar —, Oberhaunstädter — and Odenwälder ..., left and of *O. viciifolia* cv. Alvaschein —, commercial seed — and Visnovsky ..., right; symbols: solid = monocultures, open = mixtures; n = 3; bars = se.

tanniferous forage, the monocultures in this experiment yielded the better results. More work is needed to optimize mixtures and their management to produce large yields of tanniferous forage species while also being able to profit from the advantages of the mixtures such as larger total yield and weed suppression as found in our experiment and by Kirwan *et al.* (2007). It is remarkable that the important traits of yielding capacity ( $P < 0.001$ ) and proportion in mixture ( $P < 0.0001$ ) showed very important within-species variability (among cultivars of a species) (Table 1, Figure 1). Also there seems to be a certain trend for intraspecific differences in CT concentration (Table 1; Häring *et al.*, 2007b). This suggests that the potential of these species for the use as tanniferous forage plants can be further increased by plant breeding.

## Conclusions

*Onobrychis viciifolia* was the most promising CT-providing forage plant species, followed by *L. corniculatus*. The variability in competitive ability and in CT concentrations between varieties shows that there is a potential for improvement in these traits through plant breeding. Under the given management treatment, *F. pratensis* was too aggressive in mixtures with tanniferous plant species. Thus other mixtures and management techniques should be tested for the production of tanniferous forage

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# The effect of nitrogen fertilizer and harvesting frequency on yield and *in-vitro* digestibility of barley grass

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## Abstract

It is theorized that barley grass (harvested as a green crop) would benefit from nitrogen (N) fertilizer applications during early reproductive growth states. This study examined the effects of fertilizer N applications (as urea) on irrigated land at different rates and cutting intervals. It presents an assessment of our understanding of N nutrition of barley grass, with particular emphasis on use of urea-N fertilizer through the vegetative stage. The effect of different rates of N fertilizer (0, 50, 100 and 150 kg N ha<sup>-1</sup>) and harvesting frequency (2, 3, and 4-week intervals) was investigated on total dry matter (DM) production, crude protein (CP) content, *in-vitro* digestibility of organic matter (DOM) and DOM yield of the barley grass crop. The result showed that the highest rates of fertilizer application gave the greatest yield of DM and DOM and this was obtained at the longest interval between two consecutive harvesting times. In the shortest interval (15 days) between harvesting no significant ( $P > 0.05$ ) increase in yield was observed. The interaction between harvesting period and the rate of fertilizer had significant ( $P < 0.05$ ) effects on yields and DOM. The CP concentration in harvested forage showed a positive significant ( $P < 0.05$ ) correlation with fertilizer application, and DOM showed a significant ( $P < 0.05$ ) negative correlation with the length of harvesting interval.

Keywords: barley grass, urea, harvesting period, fertilizer

## Introduction

Use of nitrogen (N) fertilizer on irrigated farm land increased through the 1990s in Iran. The use of N fertilizer is now promoted and is an accepted management option to help fill the expected feed gaps on many irrigated farms. It is generally recognized that the productivity and quality of grass for hay are affected by the level of N fertilization and frequency of defoliation. It has been observed that fertilization of grass with nitrogen as urea increased dry matter (DM) and nitrogen content under irrigated condition. In this regard, increasing the clipping frequency has resulted in increased dry matter yields. It is theorized that advancing maturity is associated with declining total nitrogen content and declining digestibility of forages. The purpose of this study was to investigate the use of applications of urea fertilizer on irrigated barley grass combined with different clipping frequencies, and to determine their effects on total DM yield and nitrogen content of harvested barley grass, and on its organic matter (OM) digestibility measured by *in-vitro* methods.

## Materials and methods

The experimental design was a split-plot design with three replications. Four levels of urea (fertilizer grade, 0.46 N content) equivalent to 0, 50, 100, 150 kg N ha<sup>-1</sup>, constituted the main plots, and three clipping frequencies taken at 2, 3, and 4-week intervals were the sub-plots. The barley (as a single crop) was seeded at a rate of 70 kg ha<sup>-1</sup>. The dimensions of each unit (sub-plot) were 1.5 × 2.5 m. Urea fertilizer was applied at the end of April and clipping began in mid-May. Other conditions were the same for all main and sub-plots.

The clipped sample consisted of a 50 cm mown strip taken from the centre of each unit. The weight of the harvested fresh material was recorded, and a 500 g sample was taken from each unit and oven-dried at 60 °C to enable calculation of dry matter (DM) yield on a per-hectare basis. After drying, the samples were ground to pass through 1 mm screen and analysed for crude protein (CP) and *in-vitro* digestibility (Digestible Organic Matter, DOM). For *in-vitro* digestibility 1 g samples were incubated in diluted rumen fluid (prepared from adult sheep fed on a standard ration) for 48-h, followed by a 24-h pepsin digestion for removal of microbial protein and remaining plant protein.

Data were analysed with a model that included the effects of fertilizer level, clipping frequency and interaction. All data from each factor were analysed with the GLM procedure of SAS (2003). If a significant ( $P < 0.05$ ) main effect was detected, the main effect means were separated by a Duncan test (SAS, 2003).

## Results and discussion

The main effects of fertilizer application and clipping interval are summarized in Tables 1 and 2. Urea application had a significant effect ( $P < 0.05$ ) on all parameters at all clipping intervals (Table 1). The main effect of fertilizer on total nitrogen content in plant material was very pronounced. With each increment of urea up to 150 kg N ha<sup>-1</sup>, there was a significant increase in the concentration of crude protein. This increase in nitrogen content had no appreciable effect on the DOM, when the fertilizer rate was increased from 0 to 150 kg N ha<sup>-1</sup> (even though significant at  $P < 0.05$ ).

Table 1. Mean effect of urea application on the DM and DOM yield of barley grass and on crude protein concentration.

Fertilizer N kg N ha <sup>-1</sup>	DM yield DM (g m <sup>-2</sup> )	Digestible organic matter		Crude Protein, CP (% of DM)
		DOM (g m <sup>-2</sup> )	% of DM	
0	419d	268d	64.0c	14.9d
50	527c	322c	61.1d	16.8c
100	565b	368b	65.0a	18.6b
150	633a	409a	64.7b	20.1a
SE	3.36	2.43	0.05	0.08

Means with the same letter in each column in each section are not significantly different ( $P < 0.05$ ).  
SE: standard error.

The clipping interval (Table 2) also had a significant effect ( $P < 0.05$ ) on all criteria at all levels of urea application. Each increase in length of clipping interval gave a significant increase in yield of both DM and DOM per hectare ( $P < 0.05$ ).

Table 2. Mean effect of clipping interval on yield of barley grass.

Clipping Interval (wks)	DM (g/m <sup>2</sup> )	Digestible organic matter		Crude protein, CP (% of DM)
		DOM (g/m <sup>2</sup> )	% of DM	
2	413c	263c	63.5b	19.6a
3	547b	356b	65.0a	18.1b
4	649a	407a	62.6c	15.1c
SE	2.91	2.11	0.05	0.07

Means with the same letter in each column in each section are not significantly different ( $P < 0.05$ ).  
SE: standard error.

Increasing the interval between clippings results in a decrease in the total nitrogen content measured as crude protein concentration in the harvested DM ( $P < 0.05$ ). A sharp decline in crude protein concentration can be seen when the clipping interval advanced beyond the



fourth week stage. An increased harvested DM yield in barley grass with application of N as urea as reported here is possibly related to a faster growth of plant material. Crops that have grown quickly are usually very palatable, and that a moderate supply of available N is essential to maintain herbage growth.

The increase in harvested production as DM and DOM, obtained from treatments that had longer clipping intervals was accompanied by a sharp decline in the total nitrogen content (expressed as crude protein concentration in the harvested DM). As a result, the greatest amounts of crude protein harvested tended to be produced by barley grass mown at the second or third week stages of growth. However, it is known from the literature that responses to N fertilizer can be limited by deficiencies of other nutrients, such as phosphorus (P), potassium (K) and sulphur (S) (Whitehead, 1995). In this regard, Mundy (1993) found that addition of S with N fertilizers applied in mid-winter to a temporarily waterlogged perennial pasture had no effect on the growth response to the applied N.

There is evidence that for wheatgrass on irrigated land, clipping at 3-week intervals produced at most only 75% as much root mass as grass cut at 6-week intervals, by the end of the first harvest year. If it can be assumed that a plant with an extensive root system can make more efficient use of a heavy application of N fertilizer than a plant with less well developed roots, and this could explain why extended clipping intervals gave larger increases in DM yield at higher fertilizer rates than short clipping intervals. However, in other work, Mundy (1993) also found that frequency of defoliation can affect the cumulative growth response of a paspalum-dominant pasture to applied nitrogen in late summer.

## Conclusion

The use of urea as a source of fertilizer-N had a pronounced effect on total protein content in barley grass. This increase in yield of protein had only marginal effect on the DOM. Clipping interval also had a significant effect on the criteria measured at all levels of urea application. The increase in DM and DOM production obtained at longer clipping intervals was accompanied by a sharp decline in total crude protein concentration in the harvested DM.

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# Quality development in regrowths of timothy, meadow fescue and red clover

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## Abstract

Meadow fescue, red clover and timothy were grown in mixed swards at three sites in Norway. First cuts were taken at three different developmental stages, and succeeding regrowths sampled according to accumulated heat sum units. There was a consistent relationship between the age of regrowth and fibre digestibility and energy content. The decrease in quality by age was steeper for clover and timothy than for fescue. If high quality (in terms of digestibility and energy content) is a target, second cuts after early first cuts from these types of swards ought to be taken before the accumulation of 600 d° (base temperature 0 °C).

Keywords: digestibility, *Festuca pratensis*, NDF, *Phleum pratense*, summer growth, *Trifolium pratense*

## Introduction

The growing season in most of Norway is relatively short, lasting from 150 to 200 days with a mean temperature above 5 °C. Two cuts per season are often not sufficient to obtain a high silage quality. Research is presently undertaken to examine under what conditions harvesting regimes with an early first cut and more frequent harvests are cost-effective. Under such regimes the second and third cuts constitute a high proportion of the total yield, and knowledge of their quality potential and development will be of importance. In spring growth, observations and model simulations of the phenological stage of development of timothy may give a reliable basis for decisions about harvesting (Gustavsson *et al.*, 2003). In regrowths, the generative development may either be absent or less synchronous. Gustavsson and Martinsson (2005) argued that the standing yield is the best predictor for fibre content and digestibility of the organic matter in summer growth. In the present study we sampled regrowing swards according to accumulated heat sum units. We wanted to see to what extent this relatively simple measure gives a reliable estimate of the quality, and to gain further insight into the quality development in regrowths of the species that are the main basis for silage production in most of Norway.

## Materials and methods

Field trials with different harvesting regimes combined with two levels of N supply (120 and 240 kg N ha<sup>-1</sup>) were conducted in leys with timothy (*Phleum pratense*, var. Grindstad/Vega), meadow fescue (*Festuca pratensis*, var. Fure) and red clover (*Trifolium pratense*, var. Nordi) for three years (2004-2006). First cuts were taken from early stem elongation until early heading of timothy and second cuts according to accumulated heat sum units afterwards (base temperature 0 °C) (Tables 1-3). The intervals were determined to fit the expected length of the growing season at the three sites where the experiment was conducted. The sites were Kvit-



hamar (63°30'N; 40 m a.s.l.), (Løken (61°7'N; 550 m a.s.l.) and Særheim (58°47'N; 80 m a.s.l.). The growing season at the respective sites is 182, 149 and 209 days (mean temperature  $\geq 5^{\circ}\text{C}$ ) according to 1961-1990 normal values. At all harvests the phenological stage of development of all species according to Moore *et al.* (1991) and the total yield were recorded. The digestibility and content of ash, nitrogen, neutral detergent fibre (NDF) and indigestible NDF (INDF) in composite yield samples and samples sorted according to species were analysed by near infrared reflectance spectroscopy (NIRS) (Fystro and Lunnan, 2006). Net energy for lactation (NEL) was calculated according to Lunnan and Marum (1994). The data for each site were subjected to ANOVA with stage at first cut, age of regrowth and species (not at Særheim) as class variables (fixed effects). If the effect of species or species interactions turned out to be significant, the data sets were split up and analysed species for species with stage and age as class variables.

## Results and discussion

If quality is evaluated according to the content of INDF and NEL, meadow fescue is rated higher in regrowths than the two other species. Irrespective of stage of development at the previous cut and the age of the regrowing swards, fescue contained less INDF and more NEL (Tables 1 and 2). These differences were probably both related to the proportion of stems and organs and tissues with lignified secondary cell walls, and to principal differences in cell wall composition between grasses and legumes. Unlike timothy and red clover, meadow fescue has a low-temperature requirement for generative induction and development (Frame *et al.*, 1998; Havstad *et al.*, 2004). If all meristems induced the previous winter and spring are removed in the first cut, a meadow fescue sward will remain vegetative the rest of the growing season. Timothy and red clover develop stems and floral organs in regrowths as far as the day length requirements are met. For all species at Løken, and in the sward dominated by timothy at Særheim, the content of INDF in the regrowth also varied according to stage of development at the first cut (Tables 2 and 3). At the latest first cuts, a higher proportion of the generatively induced meristems were probably removed than at the earliest, and this resulted in a more vegetative initial regrowth. For fescue the regrowth remained vegetative, whereas new generative shoots and tillers emerged later in clover and timothy.

At harvests taken 400-500 d° after the first cut (4-5 weeks with a diurnal mean temperature of  $14^{\circ}\text{C}$ ), all species had a content of NEL well above  $6.0 \text{ MJ kg DM}^{-1}$  and an INDF-content below  $100 \text{ g kg}^{-1} \text{ DM}$  (Tables 1-2), and may be regarded as high quality forage. Second cuts taken that early constituted 20-30% of the total yield in three-cut systems. If timothy and red clover were allowed to regrow for 600 d° or more after the first cut, the content of NEL fell below  $6.0 \text{ MJ kg DM}^{-1}$  and the INDF-content was in many instances above  $100 \text{ g kg}^{-1}$  (Tables 1, 2 and 3). The second cuts constituted 40 to 60% of the total yield. If both first and second cuts were taken at early developmental stages timothy persistency was affected negatively.

Table 1. Content of NDF and INDF ( $\text{g kg}^{-1} \text{ DM}$ ) and NEL ( $\text{MJ kg DM}^{-1}$ ) in timothy, meadow fescue and red clover at Kvithamar according to accumulated heat sum units after first cut. Means for two levels of N-supply, two years (2004 and 2006) and two stages at first cut are given.

Age of regrowth	NDF			INDF			NEL		
	Timothy	Fescue	Clover	Timothy	Fescue	Clover	Timothy	Fescue	Clover
500 d°	600	580	340**	70**	50**	90*	6.1*	6.1*	6.1*
700 d°	610	600	390	120	70	120	5.5	5.7	5.6
Species effect	$P < 0.001$			$P < 0.001$			$P < 0.05$		

\*, \*\*: significant effect ( $P < 0.05$ ,  $0.01$ ) of age of regrowth within species.

Table 2. Content of NDF and INDF (g kg<sup>-1</sup> DM) and NEL (MJ kg DM<sup>-1</sup>) in timothy, meadow fescue and red clover at Løken according to developmental stage at first cut (S) and accumulated heat sum units after cutting (A). Means for two levels of N-supply and two years (2005 and 2006) are given. The level of significance for the effect of S and A is 0.05.

Stage at first cut, age of regrowth	NDF			INDF			NEL		
	Tim	Fescue	Clover	Tim	Fescue	Clover	Tim	Fescue	Clover
Stem elong., 400 d°	540	530	320	50	30	80	6.4	6.6	6.4
Stem elong., 600 d°	600	560	390	90	50	130	5.7	6.1	5.7
Early head., 400 d°	530	520	320	40	20	80	6.4	6.8	6.3
Early head., 600 d°	570	530	380	70	30	110	6.0	6.6	5.8
Effect of:	A	S, A	A	A	S, A	A	A	S, A	A
Species effect		P < 0.001			P < 0.001			P < 0.001	

Table 3. Content of NDF and INDF (g kg<sup>-1</sup> DM) and NEL (MJ kg DM<sup>-1</sup>) in regrowths of mixed swards (> 80% timothy) at Særheim. Means for 6 samples (2 replicates x 3 years, high level of N-supply) are given. ANOVA revealed significant effect (P < 0.05) of stage of development at first cut for INDF- and NEL-content and effect of age of regrowth for NDF-, INDF- and NEL-content.

Stage at first cut	Regrowth cut after 600 d°			Regrowth cut after 750 d°		
	NDF	INDF	NEL	NDF	INDF	NEL
Early stem elongation	580	90	5.7	600	130	5.3
Stem elongation	570	80	5.9	590	110	5.6
Early heading	580	80	5.9	590	110	5.5

## Conclusions

A high proportion of meadow fescue will slow the rate of decrease in quality in regrowing mixed swards containing timothy and red clover. There was a consistent relationship between accumulated heat sum units after the previous cut, and fibre digestibility and energy content in the yield. A rule of thumb might be that second cuts after first cuts taken at or before early heading of timothy, ought to be taken before 600 d° have been accumulated if high fodder quality is targeted. If followed, a side effect may be poor persistency of timothy in leys.

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# ***In-vitro* gas production of forage from semi-natural grassland as influenced by N fertilization**

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## **Abstract**

The aim of this study was to evaluate the effects of N fertilization on nutritional value of forage from semi-natural grassland, using a gas production (GP) technique. The treatments were 35, 100 and 150 kg N ha<sup>-1</sup> applied annually. Gas production profiles were fitted to a multiphasic model. Fertilization with 35 kg N ha<sup>-1</sup> did not cause changes in fermentation characteristics and organic matter digestibility (OMD) compared with fertilization with 100 and 150 kg N ha<sup>-1</sup>. Significant differences between years were observed for all GP parameters and OMD ( $P < 0.001$ ). The results indicate that reduction in N fertilization will not induce changes in forage quality regarding *in-vitro* fermentation characteristics and OMD.

Keywords: nitrogen, gas production, organic matter digestibility

## **Introduction**

The yield and quality of forage from semi-natural grasslands under extensive management may be major constraints for integrating such forage in economically viable livestock systems (Marriott *et al.*, 2004; Isselstein *et al.*, 2005). Moreover, reduction in fertilizer inputs and lower stocking rates are necessary for maintenance of semi-natural grassland to meet biodiversity and conservation targets (Marriott *et al.*, 2004), although reduction in N fertilizer application reduces yield and nutritive value of forage (Whitehead, 1995). There is a lack of information available about forage quality from semi-natural grasslands. Success of implementation more extensive management depends on further evaluation of herbage quality. The gas measuring technique has been widely used for evaluation of nutritive value of feeds. The aim of this study was to investigate effect of nitrogen fertilization on the *in-vitro* gas production characteristics of forage from semi-natural grassland grazed by sheep.

## **Materials and methods**

The field experiment was conducted during three years (2002-2004) on semi-natural grassland-association *Arrhenatheretum medioeuropaeum* (Br-BI-19), at the Faculty of Agriculture experimental field Medvednica (650 m altitude). The treatments were three fertilizer N rates applied annually (kg ha<sup>-1</sup>) - 35 (N<sub>35</sub>), 100 (N<sub>100</sub>) and 150 (N<sub>150</sub>). The treatments were replicated four times in a randomized block design. Each replicate was rotationally grazed by Charolais sheep. The details of the field experiment as well as yield and botanical composition were reported by Bošnjak *et al.* (2006). The samples were hand clipped at the beginning of each grazing period, oven dried at 60 °C for 48 h and milled to pass 1 mm sieve. For this study all samples in the same year from all grazing periods were pooled according to their relative proportions of dry matter (DM) yield in each grazing period to annual DM yield. Gas production analysis was performed on a fully automated system as described by Cone *et al.* (1996). Rumen fluid required for the incubations was obtained from four Charolais sheep fitted with a rumen fistula. Incubation was performed for 72 hours. Each sample was fermented in duplicate in two consecutive series. Gas production curves were

fitted with a three-phasic model as described by Groot *et al.* (1996) and Cone *et al.* (1997). Maximum relative rate of degradation of the non-soluble phase (Rmax2) and time at which Rmax2 is reached (tRmax2) were calculated according to Groot *et al.* (1996). *In-vitro* organic matter digestibility was calculated using equation  $OMD = 300 + 1.616 \times GP_{20} + 0.332 \times CP$  (Gosselink *et al.*, 2004), where  $GP_{20}$  is gas production after 20 h of incubation ( $ml\ g^{-1}$  organic matter (OM)) and CP is crude protein content of forage ( $g\ kg^{-1}$  DM). All data were subjected to ANOVA using SAS MIXED procedure (SAS Inst., 1996).

## Results and discussion

Asymptotic gas production in phase 1 and 2 ( $A_1$  and  $A_2$ ), as well as gas production after 72 h ( $GP_{72}$ ) did not differ significantly between N fertilizing treatments (Table 1). Furthermore, fertilization with  $35\ kg\ N\ ha^{-1}\ year^{-1}$  did not cause significant changes on fermentation kinetics compared to fertilizing with 100 and  $150\ kg\ N\ ha^{-1}\ year^{-1}$ . Cumulative gas production curves for N fertilizing treatments are shown in Figure 1.

Table 1: Gas production characteristics and OMD at different N fertilization rates.

N fertilizing treatments	A1	A2	B2	GP72	tRmax2	Rmax2	OMD
N35	60.98	168.73	8.68	280.66	10.26	0.151	739.1
N100	61.95	171.05	8.56	280.01	10.20	0.156	746.8
N150	59.22	168.00	8.56	277.12	10.13	0.153	739.8
s.e.m.	1.17	1.13	0.08	2.3	0.11	0.002	3.05
Significance	ns	ns	ns	ns	ns	ns	ns

$A_1$  – gas production ( $ml\ g^{-1}$  OM) of the soluble phase.

$A_2$  – gas production ( $ml\ g^{-1}$  OM) of the non soluble phase.

$GP_{72}$  – gas production ( $ml\ g^{-1}$  OM) after 72 h.

$B_2$  – time (h) needed to reach 50% of  $A_2$ .

Rmax2 – maximum relative rate of degradation ( $ml\ g^{-1}\ OM\ h^{-1}$ ) of the non-soluble phase.

tRmax2 – time at which Rmax2 is reached.

s.e.m. – standard error of mean.

ns – not significant ( $P > 0.05$ ).

Averaged over fertilizing treatments, significant differences between years occurred for all gas production parameters ( $P < 0.001$ ). Higher gas production in phase 1 ( $A_1$ ) occurred in 2003 ( $70.9\ ml\ g^{-1}\ OM$ ) compared with 2002 and 2004 ( $58.9$  and  $52.3\ ml\ g^{-1}\ OM$ , respectively). Gas production of the non-soluble fraction ( $A_2$ ) was significantly higher in 2003 and 2004 ( $175.8$  and  $170.4\ ml\ g^{-1}\ OM$ ) than in year 2002 ( $161.6\ ml\ g^{-1}\ OM$ ). Cumulative gas production curves averaged over N fertilizing treatments for each year are shown in Figure 1. Moreover, significantly lower  $B_2$  and tRatemax2 occurred in 2003 ( $8.14\ h$  and  $9.55\ h$ , respectively) and 2004 ( $7.99\ h$  and  $9.38\ h$ , respectively) indicated faster degradation of plant material compared with 2002 ( $9.67\ h$  and  $11.66\ h$ , respectively). N fertilization rate had no effect on organic matter digestibility (Table 1). OMD significantly varied across years ( $P < 0.001$ ). Higher OMD occurred in 2003 and 2004 ( $766.6$  and  $747.1\ g\ kg^{-1}\ OM$ ) compared to 2002 ( $712.2\ g\ kg^{-1}\ OM$ ).

Observed differences between years in gas production profiles and OMD might be caused by significant changes in botanical composition between years. In a previous paper which reports yield and botanical composition, Bošnjak *et al.* (2006) showed significant increase in annual forbs content in second (69%) and third year (45%) compared to the first year. These results are in accordance with Duru (1997) and Lopez *et al.* (1991) who reported that dicots have faster *in-situ* degradation rates and may have higher digestibility than grasses of the same age.

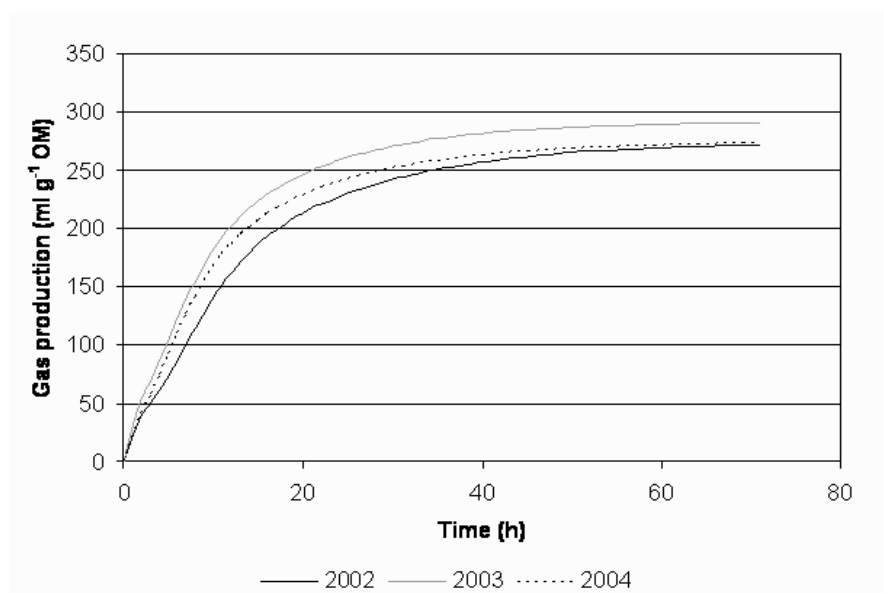


Figure 1. Cumulative gas production of forage in different years.

## Conclusions

It can be concluded that estimated fermentation parameters are not influenced by N fertilization.

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## Stability of fatty acids in grass silages after exposure to air

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### Abstract

Knowledge on the fate of fatty acids (FA) in silages that are exposed to air is scarce. During the feedout phase, oxidation might lower the concentration of unsaturated FA. Therefore, the aim was to obtain insight into changes in FA after opening of the silages.

Four grass silages with contrasting ammonia and pH values were examined in the anaerobic situation as well as during the aerobic phase after 8, 16 and 24 h of exposure to air. Samples were collected and transported in cooled plastic bags with a N<sub>2</sub> atmosphere, stored at -18 °C for 5-6 days, freeze-dried, milled, and FA were extracted with chloroform/methanol. FA concentrations were determined by gas chromatography. The FA content and composition differed greatly between the investigated silages. This study showed that exposure to air up to 24 h did not affect the fatty acid content or composition of the ensiled forage.

Keywords: silage, fatty acids, oxidation

### Introduction

In the last decades, concerns have been raised about fatty acids in human diets in relation to human health. To produce milk with a relatively high proportion of (poly) unsaturated fatty acids, animals need a dietary source of these fatty acids. In Western Europe, the indoor ration of high-producing dairy cows mainly contains ensiled grass and maize, next to concentrates. It is known that the (un)saturated fatty acid contents of herbage can be reduced through oxidation and lipolysis during conservation processes such as wilting and ensiling. After the first phase of the ensilage process, provided that compaction and sealing are sufficient, chemical processes in the silage are minimal. At the moment of opening the silage, during the feedout phase, this may change. Parts of the silage will then be exposed to air, introducing the possibility of oxidation again. This may have practical implications, as not all silage is consumed directly after having been offered to animals. The aim of this pilot study was to obtain insight into whether changes in the fatty acid concentration and composition occur after opening of the silages.

### Materials and methods

Four grass silages were used (Table 1) that had been harvested from May (first cut) to September 2006 (fourth cut). These silages had been sampled 6-8 weeks after ensiling by a commercial laboratory (Blgg, Oosterbeek, The Netherlands) and were chosen because of differences in pH and ammonia content. For this study, samples of the silages were taken by an experienced employee of Blgg on 2 and 3 May 2007 from the middle (in height) at the end of the silage clamps with a hollow drill (2 cm diameter). Samples (> 100 g) were immediately stored in plastic bags, flushed with N<sub>2</sub> and stored in a cool box (4 °C) filled with N<sub>2</sub> during transport to the laboratory, which took less than 4 hours. In the laboratory, the samples from each silage were thoroughly mixed and three subsamples were taken. One subsample was immediately stored at -20 °C in a plastic bag with a N<sub>2</sub> environment (control sample, t = 0) and the other two subsamples were spread out at room temperature to mimic the situation during the feedout phase on farms. One subsample was exposed to air for 8 h (silage 4) or 16 h

(silages 1, 2 and 3) and one for 24 h (silages 1-4,  $t = 24$ ). Each of these treatments was done in duplicate. After exposure to air, the samples were stored in the freezer at  $-18^{\circ}\text{C}$  in a  $\text{N}_2$  environment. Five to six days later, samples were freeze-dried, ground at 1 mm and stored in glass bottles with  $\text{N}_2$  at  $4^{\circ}\text{C}$  in the dark. After six weeks, lipids were extracted with chloroform/methanol (Folch *et al.*, 1957) and transesterified to FA methyl esters that were quantified using gas chromatography.

Data were analysed in SPSS (SPSS for Windows, Release 15.0.0, 2006. SPSS Inc., Chicago, IC, USA) averaging the four silage data to a mean value for each time point. The statistical model was  $Y_{ij} = M + T_i + E_{ij}$ , where  $Y_{ij}$  is the observation;  $M$  is the overall mean;  $T_i$  is the time effect ( $i = 0, 1, 2$ ); and  $E_{ij}$  is the residual effort. A significance level of 0.05 was used.

Table 1. Harvest date, cut number, dry matter (DM) content ( $\text{g kg}^{-1}$ ), and pH and  $\text{NH}_3$  content of 4 silages 6-8 weeks after ensiling.

Silage	Harvest	Cut	DM	pH	$\text{NH}_3$
A	June	2	525	high	high
B	Sept	4	346	low	high
C	May	1	557	high	low
D	July	3	300	low	low

## Results and discussion

There were large differences in concentrations of total FA and the main individual FA (C16:0, C18:2, C18:3) between the different grass silages (Table 2). The FA content in silages C and D was 2 to 3 times higher than in silage B. Similar differences were observed for individual FA. These differences may be related to the time of harvest, days of regrowth, age of harvested grass, species composition, N fertilization, seasonal factors, management strategies, fermentation processes, etc.

Table 2. Concentrations of dry matter (DM), C16:0, C18:2, C18:3 and total fatty acids (FA) in four anaerobic silages ( $t = 0$ ) and after 8 or 16 h, and 24 h of exposure to air.

Silage	Time	DM ( $\text{g kg}^{-1}$ )	C16:0 ( $\text{g kg}^{-1}$ DM)	C18:2 ( $\text{g kg}^{-1}$ DM)	C18:3 ( $\text{g kg}^{-1}$ DM)	Total FA ( $\text{g kg}^{-1}$ DM)
A	0	583	3.4	2.5	11.2	18.2
	16	621	3.5	2.5	11.1	18.1
	24	632	3.5	2.6	11.5	18.6
B	0	518	2.7	2.1	4.0	9.4
	16	585	2.5	2.0	3.7	8.9
	24	597	2.5	2.0	3.7	8.8
C	0	639	3.6	2.9	13.8	21.5
	16	671	3.7	2.9	13.8	21.7
	24	689	3.7	3.0	14.2	22.1
D	0	488	4.1	4.0	11.8	21.3
	8	504	4.0	3.9	11.6	20.8
	24	549	4.0	3.8	11.2	20.4

The contents of fatty acids and concentrations and proportions of the three major individual fatty acids did not change after exposure to air up to 24 hours (Tables 2 and 3). Therefore, the hypothesis that oxidative losses of fatty acids might occur after opening of the silage due to exposure to air during the feedout phase, was not confirmed. During the ensiling phase, neither Chow *et al.* (2004), nor Steele and Noble (1983) observed a reduction in the concentration of total fatty acids (reviewed by Dewhurst *et al.*, 2006).



Table 3. Concentrations of DM, C16:0, C18:2, C18:3 and total fatty acids (FA) and proportions of FA averaged for four silages, and significance of effects of exposure to air for 0 (t = 0), 8 or 16 (t = 8-16) or 24 h (t = 24).

Time	DM (g kg <sup>-1</sup> )	C16:0 (g kg <sup>-1</sup> DM)	C18:2 (g kg <sup>-1</sup> DM)	C18:3 (g kg <sup>-1</sup> DM)	Total (g kg <sup>-1</sup> DM)	C16:0 %	C18:2 %	C18:3 %
0	557	3.5	2.9	10.2	17.6	21	17	56
8-16	595	3.4	2.8	10.0	17.4	21	17	56
24	617	3.4	2.8	10.2	17.5	21	17	56
F	23	0.08	1.47	0.50	0.72	1.24	0.11	0.60
P	0.001	0.93	0.30	0.63	0.52	0.36	0.91	0.58
SEM	6.2	0.03	0.03	0.13	0.16	1	1	2

Additional research should therefore focus on understanding the origin of differences in concentrations of fatty acids between silages, in order to provide conserved forage that could contribute to the production of 'healthier' milk with higher levels of unsaturated fatty acids. It will be necessary to develop more knowledge about grasses used for silage-making, i.e., effects of regrowth days, N fertilization, time of cut, seasonal factors, and species (reviewed by Elgersma *et al.*, 2006).

## Conclusions

The fatty acid content in grass silages and the concentrations and proportions of the three major individual fatty acids did not change after opening and exposure to air up to 24 hours.

## Acknowledgements

We thank Christie Wever for technical assistance.

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## Preservatives and machines with knives improve the silage quality in round bales

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### Abstract

In a dairy extension project aimed at improved silage quality more than 1100 silage samples from the harvests 2001-2006 were tested. Almost 600 farmers contributed samples, each farmer received a free analysis if details about the harvesting and storing technique accompanied the fresh silage. 45% of the silage samples came from round bales. Signs of spore-forming *Clostridia* bacteria as butyric acid were found in 45% of the samples cut using machines without knives and made without use of preservatives. In silage from machines with knives but without use of preservatives the frequency of *Clostridia* fermentation was reduced by one-third. In silage from machines with knives and use of preservatives the frequency with butyric acid was reduced by another third. Increased frequencies of butyric acid fermentation with increasing time between baling and wrapping with plastic were also found, especially if no preservatives were used, and with increasing clover content and with decreasing DM. Butyric acid was often accompanied by increased crude protein content bound to acid detergent fibres, indicating heating processes before anaerobic conditions had been established.

Keywords: preservatives, butyric acid fermentation, round bale silage

### Introduction

Cheese production is very important for the Norrmejerier Dairy. In order to minimize clostridial fermentation during the cheese storage the spore content in the milk is recorded. Depending on the frequency and degree of spore contamination the farmers are getting warnings or reduced payment for the milk. In spite of this, the economic losses arising from discarded cheese were about 2 million SEK in 2000 and 2001. The escalating frequency of spore-contaminated milk, from 15% of the dairy farms during 1995 to 50% in the year 2000, could not be explained by differences in weather conditions. A survey was therefore initiated to look at different factors that can affect the silage quality at farm level.

### Materials and methods

Milk producing farmers were offered free analysis if the delivered fresh silage sample was accompanied with details about the harvesting and storing technique. A 50g sample of the fresh silage was mixed with 750g deionised water. After trickling through four layers of cheesecloth, tubes with the diluted silage juice were sent together with dried and milled parts of the silage samples to Dairy One (Ithaca, NY, USA) for analysis according to the Cornell Net Carbohydrate and Protein System (CNCPS) (Sniffen *et al.*, 1992). Of the preservatives used in round bales, 13% were based on formic + propionic acids, 33% were inoculants and 23% were Kofasil Ultra. Statistical analysis was made in Excel. Results of the analysis were commented on and sent to the farmers together with suggestions for possible improvements.

## Results and discussion

From the harvests 2001-2006 more than 1,100 silage samples from almost 600 farmers were analysed. 45% of the samples come from farms with round bales and 55% from bunker or upright silos. Clostridial fermentation was recorded as butyric acid in 29% and as iso-butyric acid in 62% of all silage samples. Preservatives were used in > 80% of samples from silos but in *ca.* 25% of analysed round bales. Table 1 summarizes the results of this analysis. Crude protein (CP) varied from < 70 g CP kg<sup>-1</sup> DM to > 240 g CP kg<sup>-1</sup> DM, and DM content ranged from 19 to 74%. This indicates a very big variation in maturity stage and weather conditions during the harvest, resulting in variation in the extent and pattern of fermentation (Table 2). The water soluble carbohydrates varied from zero to almost 28% of the DM.

Table 1. Variation in silage quality and nutrient concentrations in tested round bale silage samples.

	DM, %	ME MJ kg <sup>-1</sup>	Crude Protein g kg <sup>-1</sup>	NDF g kg <sup>-1</sup>	Ca g kg <sup>-1</sup>	P g kg <sup>-1</sup>	Mg g kg <sup>-1</sup>	K g kg <sup>-1</sup>	pH
Mean	38	10.5	141	555	7.0	2.7	1.9	20.2	4.2
St. dev	12	1.0	32	61	2.8	0.7	0.5	5.9	0.4
Max.	74	13.3	246	735	19.2	5.2	5.1	42.5	6.1
Min.	19	6.7	66	289	1.4	0.6	0.5	5.6	3.3

Table 2. Concentrations of acids and ammonium N in round bale silage DM and proportion of samples with traces of spore-forming *Clostridia* bacteria as butyric acid and iso-butyric acid.

	Lactic acid g kg <sup>-1</sup>	Acetic acid g kg <sup>-1</sup>	Propionic acid g kg <sup>-1</sup>	Butyric acid g kg <sup>-1</sup>	Isobutyric acid g kg <sup>-1</sup>	Frequency Butyric acid	Frequency Isobutyric acid	NH <sub>4</sub> N as % of total N
Mean	44.9	17.0	0.7	0.6	1.1	32%	67%	6.0
St. dev	23	8.4	1.8	2.0	1.9			3.6
Max.	116.9	49.5	14.4	21.7	21.2			27.6
Min.	0	1	0	0	0			0

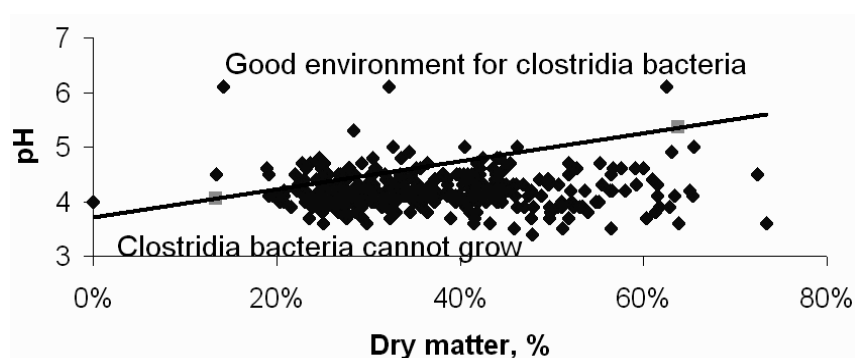


Figure 1. Round bales with butyric acid plotted against actual pH and dry matter content.

Traces of spore-forming *Clostridia* bacteria as butyric acid and iso-butyric acid have been found in silage up to 70% DM. In Figure 1, round bales with butyric acid are plotted against the actual silage DM and pH. In environments below the plotted line,  $((0.0257 \cdot \text{pH} \cdot 100) + 3.75)$ , *Clostridia* bacteria do not grow (Svensk Mjöljk, 2000). Figure 1 shows that most of the silage with butyric acid has a pH below that at which *Clostridia* can thrive. The reasons why they

still have been able to grow must, therefore, be due to factors contributing to a too slow fermentation process.

A test of the spore content in milk and silage among farmers who delivered milk to the Burträsk cheese plant, showed higher correlation between the spore content in milk and the amount of butyric acid in fed silage ( $R^2 = 0.55$ ) than between the spore content in milk and the spore content in the silage ( $R^2 = 0.13$ ). This is a sign that *Clostridia* can change to spores during the passage through the intestine. The correlation between spores in milk and iso-butyric acid in silage was lower ( $R^2 = 0.25$ ). As shown in Figure 2a, butyric acid was found in 45% of the samples of silage cut using baling machines without knives and made without preservatives. The butyric acid frequency was reduced, with one-third in silage cut using machines with knives but without preservatives. This frequency was reduced by another third in round bales from machines with knives and with use of preservatives. An earlier harvest may also have contributed to this; the mean (and sd) values were CP 163 (38) g kg<sup>-1</sup> and NDF 516 (62), compared with CP of 133 (28) g kg<sup>-1</sup> DM, and NDF of 566 (56) g kg<sup>-1</sup> DM in untreated silage. DM content was in the same range for all groups, mean (and sd) 36-38% (10-12). Provision of a fix or flex press chamber to the baling machine does not seem to matter. Figure 2b shows that the risk for clostridial fermentation seems to increase with the length of time between baling and wrapping with plastic if no preservatives are used. Often butyric acid is found together with increased levels of protein bound to ADF. This is a sign of aerobic heating processes before plastic wrapping and oxygen removal and something that may have encouraged the *Clostridia* bacteria to grow. The lower frequency of butyric acid in silage with preservatives may be an effect of a more rapid drop in pH and less heating. The higher frequency of butyric acid in treated silage wrapped 1-2 h after baling may be due to much higher clover content and perhaps, therefore, of too low an application rate of the preservative. Growth of *Clostridia* bacteria seem to result in protein breakdown, as the results of the analysis show increased levels of ammonium and other soluble protein fractions together with the butyric acid.

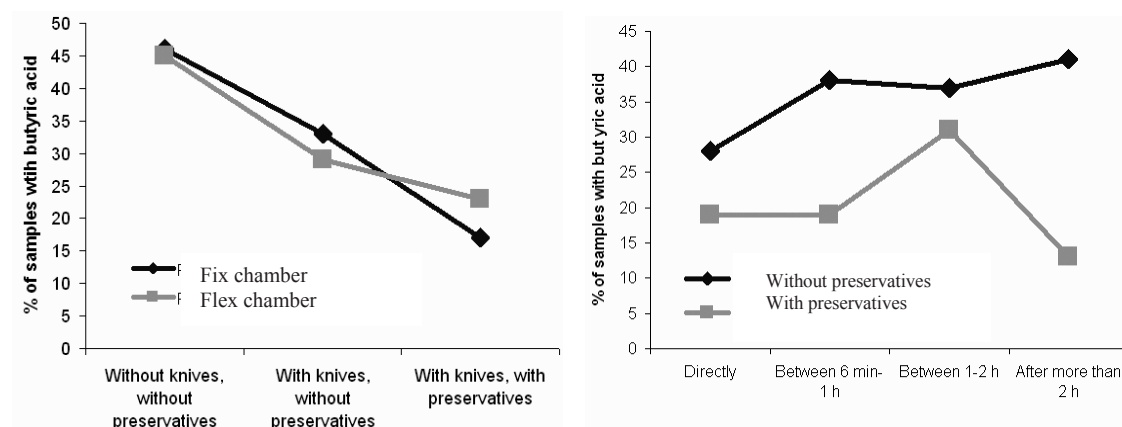


Figure 2. a) Effects of preservatives and type of baling machine on the frequency of butyric acid. b) Effects of time between baling and wrapping with plastic and preservatives on the butyric acid frequency in round bale silage.

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# Digestibility and fibre content of leaves and straw of three *Festulolium* hybrids during spring regrowth

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## Abstract

Three *Festulolium* hybrids (x *Festulolium* spp.), namely Paulita and Perun (*Lolium multiflorum* x *Festuca pratensis*) and Hykor (*Lolium multiflorum* x *Festuca arundinacea*) were sampled during spring regrowth. Samples were taken three times during a period that covered stages from early to late reproductive development. These were cut at ground level within each of the three replicate plots. The samples were mixed and a random sub-sample was separated into leaf and straw material. The samples were dried at 55 °C for 30h, weighed, milled and analysed for organic matter (OM) digestibility, neutral detergent fibre (NDF) and insoluble neutral detergent fibre (iNDF). OM and NDF were analysed chemically while iNDF was analysed by near reflectance spectroscopy (NIRS). Hykor had a significantly greater leaf proportion at all harvest occasion than Paulita and Perun. Leaf digestibility of Hykor differed little with phenological stage, while that of Paulita and Perun declined with time. Straw digestibility declined with phenological development and that of Hykor was generally poorer than that of Paulita and Perun. The content of NDF and iNDF in both leaf and straw material was significantly greater at all harvest occasions for Hykor compared with Paulita and Perun.

Keywords: *Festulolium*, digestibility, NDF, iNDF, reproductive growth

## Introduction

New high-yielding hybrid-grasses have recently been introduced on the Swedish seed market. To be useful for the Swedish farmer, a grass species must not only be high-yielding but also meet high demands for nutritive value, particularly digestibility. The aim of this study was to evaluate three recently introduced *Festulolium* hybrids (x *Festulolium* spp.) with respect to the development of their digestibility and fibre content during spring regrowth.

## Materials and methods

Three *Festulolium* hybrids, namely Paulita and Perun (*Lolium multiflorum* x *Festuca pratensis*) and Hykor (*Lolium multiflorum* x *Festuca arundinacea*) were sampled three times (25 May, 2 and 7 June 2007) during a period that covered stages from early to late reproductive development. The plots were part of a field trial which was established for the official variety testing of ryegrasses at Rådde (57°36'N, 13°16'E) in spring 2005. The plots received 28 kg P ha<sup>-1</sup>, 100 kg K ha<sup>-1</sup>, 15 kg S ha<sup>-1</sup> and 100 kg N ha<sup>-1</sup> 17 April 2007. The experimental lay-out was a completely randomized block design with three replicates. The mean daily temperature during the period studied increased from 12 to 20 °C, and the plots never suffered from drought.

Four samples, each with an area of 0.03 m<sup>2</sup>, were cut at ground level within each of the plots. The samples were mixed, and a random sub-sample was separated into leaf and straw (including leaf sheaths) material. These samples were dried at 55 °C for 30h, weighed, milled and analysed for organic matter (OM) digestibility, neutral detergent fibre (NDF) and insoluble neutral detergent fibre (iNDF). OM and NDF were analysed chemically while iNDF was analysed by near reflectance spectroscopy (NIRS).

The data were analysed as a repeated measures analysis using the MIXED procedure in SAS/Stat software, version 9.1 of the SAS Systat for Windows.

## Results

Hykor had a significantly larger leaf proportion than Paulita and Perun on all harvest occasions ( $P < 0.001$ , Table 1). The proportion declined with time, and more for Paulita and Perun ( $P < 0.05$ ).

Table 1. Metabolizable energy content and leaf proportion of three festulolium hybrids during spring regrowth.

Variety	Leaf energy content			Straw energy content			Leaf proportion		
	MJ kg DM <sup>-1</sup>			MJ kg DM <sup>-1</sup>					
	25 May	2 June	7 June	25 May	2 June	7 June	25 May	2 June	7 June
Hykor	11.4a*	11.3a	11.2a	11.4a	10.8a	9.5a	0.63a	0.58a	0.39a
Paulita	11.4a	11.1a	10.0a	12.5b	11.6b	10.8b	0.42b	0.28b	0.20b
Perun	11.7a	11.3a	10.6a	12.6b	11.5b	10.9b	0.44b	0.31b	0.21b

\*Different letters indicate significant differences between cultivars.

The digestible energy content of the leaves did not differ between the three cultivars or between the two first harvest occasions, but thereafter Paulita and Perun lost some of their energy content ( $P < 0.05$ , Table 1). The digestible energy content of the straw was generally lower for Hykor than for Paulita and Perun ( $P < 0.001$ ) and it generally decreased progressively with time ( $P < 0.001$ ). The NDF content of both leaves and straw was generally larger for Hykor than for Paulita and Perun ( $P < 0.001$ , Table 2). The NDF content of the leaves increased somewhat with time for all varieties ( $P < 0.05$ ). The NDF content of the straw material also increased with time ( $P < 0.001$ ), but more for Paulita and Perun than for Hykor ( $P < 0.01$ ).

Table 2. Fibre content (NDF) of three festulolium hybrids during spring regrowth.

Variety	Leaf NDF content g kg DM <sup>-1</sup>			Straw NDF content g kg DM <sup>-1</sup>		
	25 May	2 June	7 June	25 May	2 June	7 June
Hykor	563a*	565a	589a	658a	683a	724a
Paulita	450b	460b	465b	538b	621b	672b
Perun	450b	465b	485b	529b	623b	683b

\*Different letters indicate significant differences between cultivars.

The iNDF content of both leaves and straw was generally larger for Hykor than for Paulita and Perun ( $P < 0.001$ ), particularly for the leaves (Table 3). The content of the leaves increased somewhat with time ( $P < 0.001$ ), and more for the straw ( $P < 0.001$ ).

Table 3. Indigestible fibre content (iNDF) of three festulolium hybrids during spring regrowth.

Variety	Leaf iNDF content g kg DM <sup>-1</sup>			Straw iNDF content g kg DM <sup>-1</sup>		
	25 May	2 June	7 June	25 May	2 June	7 June
Hykor	61a*	64a	78a	93a	106a	150a
Paulita	13b	14b	19b	53b	87b	121b
Perun	18b	12b	26b	61b	88b	131b

\*Different letters indicate significant differences between cultivars.

Whole crop content of the respective variables can be calculated from the data presented, and these results (not presented) show that Hykor generally had a smaller energy content ( $P < 0.01$ ).



and a larger NDF ( $P < 0.001$ ) and iNDF ( $P < 0.001$ ) content than either Paulita or Perun. The latter two did not differ significantly in any variable.

## Discussion

The genetic background of the hybrids obviously played a major role in the determination of the fibre content and digestibility of their leaves and straw. Thus, Paulita and Perun, which had *Festuca pratensis* as one of their parents, showed identical quality traits. It is interesting to note that in these hybrids the straw had a greater energy content than the leaves throughout the period studied, which is unusual. Hykor, with *Festuca arundinacea* as one parent, differed significantly from Paulita and Perun as regards leafiness and most quality traits. Leafiness is usually associated with high herbage quality (e.g. Kalu *et al.*, 1988). However, this study shows that even though Hykor maintained a high leaf proportion as reproductive development advanced this did not compensate for the decrease in the energy content of the straw, and the energy content of the whole crop was always poorer with Hykor than with Paulita or Perun. Tall fescue is characterized by its rough leaves, and is known to have a poor digestibility compared with other grasses (e.g. Demarquilly and Jarrige, 1973). However, Pozdisek *et al.* (2003) found that Hykor was an improvement over *Festuca arundinacea* as regards digestibility, while Ghesquiere *et al.* (1996) found that hybrids of *Lolium multiflorum* x *Festuca arundinacea* were similar in palatability to the respective parent species. We conclude that Hykor must be harvested earlier than either Paulita or Perun if a high digestibility of the crop is of high priority.

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# Quality of forage maize (*Zea mays* L.) depending on sowing time in Latvia

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## Abstract

Despite the southern origin of maize (*Zea mays* L.), this crop is used for forage more widely every year in Latvia. For high-quality silage making, sowing time is important. Field trials with four maize hybrids ('Earlstar', 'RM-20', 'Tango', 'Cefran') sown on four dates (April 25, May 5, May 15, May 25) during 2005-2007 were carried out at Vecauce, Latvia. Maize was harvested on four dates beginning on 1 September and at 10-day intervals thereafter. Results showed that sowing time had a slight (3 to 7%), but significant ( $P < 0.05$ ) effect on dry matter (DM), crude protein, neutral and acid detergent fibre contents, and the proportion of ears in the whole DM yield. Our preliminary conclusion is that the best sowing time in central and western Latvia is near May 5. Maize sown earlier (April 25) or later (May 15) also performs well, but performance at these sowing dates depends on the specific year. The sowing time effect was more pronounced in years when the growth and development of plants occurred normally (2007) or when autumn frosts were observed earlier in September (2005).

Keywords: forage maize, sowing date, yield, quality, dry matter, ear proportion

## Introduction

Maize (*Zea mays* L.) is mainly grown between latitudes 30° and 55°, and relatively little is grown at latitudes higher than 47°. Despite Latvia being located between latitudes 55° and 58°N, maize is used for forage more widely every year. For high-quality silage making in such a marginal area every step of the growing process is important. Much research on sowing time in maize has been carried out in other agro-meteorological conditions, mainly taking into account the grain but not silage yield (Lauer *et al.*, 1999; Racz *et al.*, 2003). In most cases earlier sowing had a positive effect on maize yield. Rossman and Cook (1966) (cited in Racz *et al.*, 2003) observed in an experiment carried out over a 10-day period that the grain yield was reduced by 9% when sowing was delayed by 10 days. Gaile (2004) reported that dry matter (DM) yield of the whole maize plant in Latvia during four years was reduced by 7 to 12% when sowing was delayed by 10 days after the optimum. Gaile (2004) determined that early planting depends on conditions of the particular season, mainly air temperature, and is one of the keys to growing silage maize in Latvia. However, this research did not specify the optimum sowing date. It only indicated that a delay in planting can cause reductions in yield along with reduced DM content and density of ear portion in the whole-plant DM yield at harvesting. These factors, along with nutritive value measures such as content of neutral detergent fibre (NDF), acid detergent fibre (ADF), crude protein (CP) and net energy for lactation (NE<sub>L</sub>) are very important, especially when we prepare diets for highly productive dairy cows.

The aim of the research described in this paper was to analyse maize quality as affected by sowing time, and to define more accurately the optimum sowing time for maize in Latvia. The hypothesis was that sowing maize earlier (very late April to early May) will ensure higher yields with an improved nutritive value.

## Materials and methods

Three-factor field trials were carried out from 2005 to 2007 at the Research and Study farm Vecauce (latitude 56°28'N, longitude 22°53'E) of Latvia University of Agriculture. Trials were arranged in a randomized complete block design with four blocks with an individual plot size of 16.8 m<sup>2</sup>. Planted population density was 82,000 plants ha<sup>-1</sup>. Seed of four maize hybrids (Factor A) with different maturity ratings defined by the FAO number was used (Table 1). Soil at the site was sod podzolic sand loam with pH<sub>KCl</sub> of 7.0-7.1, an available P content of 198-263 mg kg<sup>-1</sup>, available K of 191-196 mg kg<sup>-1</sup>, and a humus content of 21-25 g kg<sup>-1</sup>. Maize was sown on four dates (Factor B) starting on April 25 and at ten-day intervals until May 25 of each year (Table 1). Traditional soil tillage was used, which included mould-board ploughing in the previous autumn along with cultivation and rotor-tilling before sowing in spring. Fertilizers applied were 34 kg P ha<sup>-1</sup>, 75 kg K ha<sup>-1</sup> and 148 kg N ha<sup>-1</sup> (18 + 70 + 60). Weeds were controlled mechanically and with herbicides. Harvesting was done on four dates (Factor C; results not yet available) beginning on 1 September and continuing at ten-day intervals. In this paper results are presented for the DM yield, proportion of ear DM yield in the whole plant DM yield, content of DM of whole plant and ears, CP, NDF and ADF. Net energy for lactation was calculated as: NE<sub>L</sub>, MJ kg<sup>-1</sup> of DM = (0.00245 × DDM - 0.12) × 4.184, where DDM is digestible dry matter. Meteorological conditions varied between the research years, but all three years were, on average, suitable for maize growing. Average day and night temperature from 25 April to 30 September was 14.4 °C in 2005, 15.7 °C in 2006, and 14.8 °C in 2007. Total precipitation over the same period and years was 298, 267 mm, and 339 mm, respectively. A spring frost after maize emergence occurred on 1 June 2006. Strong autumn frosts occurred on 17 and 18 September 2005 when maize was heavily damaged. Hazardous frosts were not observed in 2007. Results were statistically analysed using standard analysis of variance methods for a 4 × 4 × 4 factorial arrangement of treatments in a randomized complete block design.

## Results and discussion

The largest average three-year DM yield was obtained when maize was sown on May 5 (14.81 Mg ha<sup>-1</sup>). Sowing date had a small (7.5%,  $P < 0.05$ ) but significant effect on average DM yield per trial period. This effect was more pronounced in 2005 (19%,  $P < 0.05$ ) and in 2007 (16%,  $P < 0.05$ ). The first silage quality indicators for maize over the years is the DM content in the whole plant yield (at least 250 g kg<sup>-1</sup>) and the proportion of ear yield (at least 400 g kg<sup>-1</sup>) in the whole plant DM yield. Preliminary research (Gaile, 2004) suggested that maize sown earlier can achieve improved quality at harvest. Data from the current study showed that substantially higher DM content was obtained when maize was sown on April 25 compared with the other sowing dates ( $P < 0.05$ ) (Table 1). Similarly, substantially higher ear proportion was observed when maize was sown in the first three sowing dates (up to May 15; Table 1). The same was related to average three-year values for NE<sub>L</sub> (6.49 MJ kg<sup>-1</sup> on April 25, 6.46 MJ kg<sup>-1</sup> on May 5, 6.44 MJ kg<sup>-1</sup> on May 15, and 6.38 MJ kg<sup>-1</sup> on May 25). Sowing date also had small, but substantial ( $P < 0.05$ ) effects on such indicators as average three-year NDF (2.6%) and ADF contents (3.3%): they increased with sowing delay in May. Since maize is not a typical plant for the Latvian climate, all the described parameters were substantially affected by meteorological conditions during the trial years. NE<sub>L</sub> and contents of NDF and ADF depended on it most of all: in the warmest year (2006) the average per trial NE<sub>L</sub> was highest (6.66 MJ kg<sup>-1</sup>), but the contents of NDF (491 g kg<sup>-1</sup>) and ADF (243 g kg<sup>-1</sup>) were lowest.

Table 1. Dry matter content of maize and proportion of ears in the whole plant DM yield ( $\text{g kg}^{-1}$ ) as affected by hybrid and sowing date in Latvia.

Harvest dates	Hybrid				Averages for sowing dates
	Earlstar FAO-160	RM-20 FAO-180	Tango FAO-210	Cefran FAO-340	
DM content ( $P > 0.05$ for hybrid x sowing date)					$\text{LSD}_{0.05} = 7.3, P < 0.05$
April 25	296	280	291	241	277
May 5	284	275	281	235	269
May 15	271	266	271	227	259
May 25	265	247	252	217	245
Averages for hybrids, $\text{LSD}_{0.05} = 7.3, P < 0.05$	279	267	274	230	
Proportion of ear DM yield in the whole plant DM yield ( $P > 0.05$ for hybrid x sowing date)					$\text{LSD}_{0.05} = 21.5, P < 0.05$
April 25	512	510	532	403	489
May 5	500	480	534	395	477
May 15	532	507	527	377	486
May 25	466	423	465	296	412
Averages for hybrids, $\text{LSD}_{0.05} = 21.5, P < 0.05$	503	480	514	368	

Only CP content improved with delayed sowing. Because maize is used mainly to provide energy in dairy cow diets, the decrease in CP when maize is sown earlier can be tolerated.

## Conclusions

The best time to sow maize in the central and western part of Latvia is near May 5. This sowing date provided the highest DM yield and the quality was of equal value to maize sown on April 25. Maize sown earlier (April 25) or later (May 15) also performs well, but performance depends on the specific year, and on the parameter evaluated. A greater effect of sowing time on yield and quality was observed in years when the growth and development of plants occurred normally (2007) or when autumn frosts were observed earlier in September (2005). If sowing is delayed up to the end of May, only very early hybrids (such as Earlstar, FAO 160) can provide good quality yield. Late hybrids (such as Cefran, FAO 340) are not suitable for Latvian conditions at all, even when sown early.

## Aknowledgements

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# Changes in feed quality of different forage legumes during autumn growth

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## Abstract

In autumn growth forage legumes dominate, resulting in high contents of crude protein (CP) in the sward. The aim of the present study was to determine the variation in feed quality of white clover, red clover, kura clover, alfalfa and birdsfoot trefoil harvested on three different dates (approximately 22, 46 and 60 days of regrowth after 5 August) in three years. Swards were established each year as binary swards with perennial ryegrass. CP, CP-fractionation, neutral detergent fibre (NDF), acid detergent fibre (ADF) and energy contents were measured and energy content estimated. Results show a significant species x year x sampling date interaction for the analysed forage quality parameters. Highest CP values were achieved by white clover, alfalfa and red clover with 30% in the early cut. The highest content of fraction A (non-protein N) was also found on this sampling date, decreasing in the later harvests. Red clover had the lowest contents of fraction A and, together with birdsfoot trefoil, the highest contents of fraction C (in ADF-residue-bound N = acid detergent insoluble N). Considering N use efficiency in ruminant nutrition, red clover and birdsfoot trefoil seem to have positive effects, compared with other forage legumes, on effective N use.

Keywords: legumes, autumn growth, forage quality, protein

## Introduction

In north-west Europe, white clover plays an important role in grassland systems. Forage legume-based swards usually have a high proportion of legumes in autumn. Crude protein (CP) content in such swards is proportional to the legume content in the harvestable biomass. This protein may be poorly used by ruminants as it contains large amounts of non-protein N, which leads to low N-use efficiency by the animals (Beever *et al.*, 1986). However, differences in legumes in terms of content of secondary plant compounds may be advantageous to avoid excessive N losses in production systems. Secondary compounds of relevance for improved N utilization in animal are the condensed tannins found in birdsfoot trefoil and the polyphenol oxidase found in red clover. The aim of the present study was to measure the variation in forage quality of different forage legumes in the autumn regrowth, to find proper harvest management, taking into consideration forage legume species and their protein quality.

## Material and methods

Binary mixtures with perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*), red clover (*Trifolium pratense*), alfalfa (*Medicago sativa*), birdsfoot trefoil (*Lotus corniculatus*) or kura clover (*Trifolium ambiguum*) were established yearly in the autumn, and sampling for this experiment was performed in the first production year of three consecutive years. In spring, plots were treated equally with a late first-cut at the end of June. A uniform cut was performed at the beginning of August of each year and samples were collected at approximately 22, 46 and 60 days afterwards, cutting at 5 cm height. The legume proportion was determined on a dry matter (DM) basis by separating legume, grass and weeds. The quality analyses were performed on the legume component only. For each legume species,

CP, NDF, ADF, NEL and CP-fractionation (Licitra *et al.*, 1996) were determined. Data were submitted to analysis of variance and means were compared to white clover using Student's t-test. Probabilities were adjusted using the Bonferroni-Holm test.

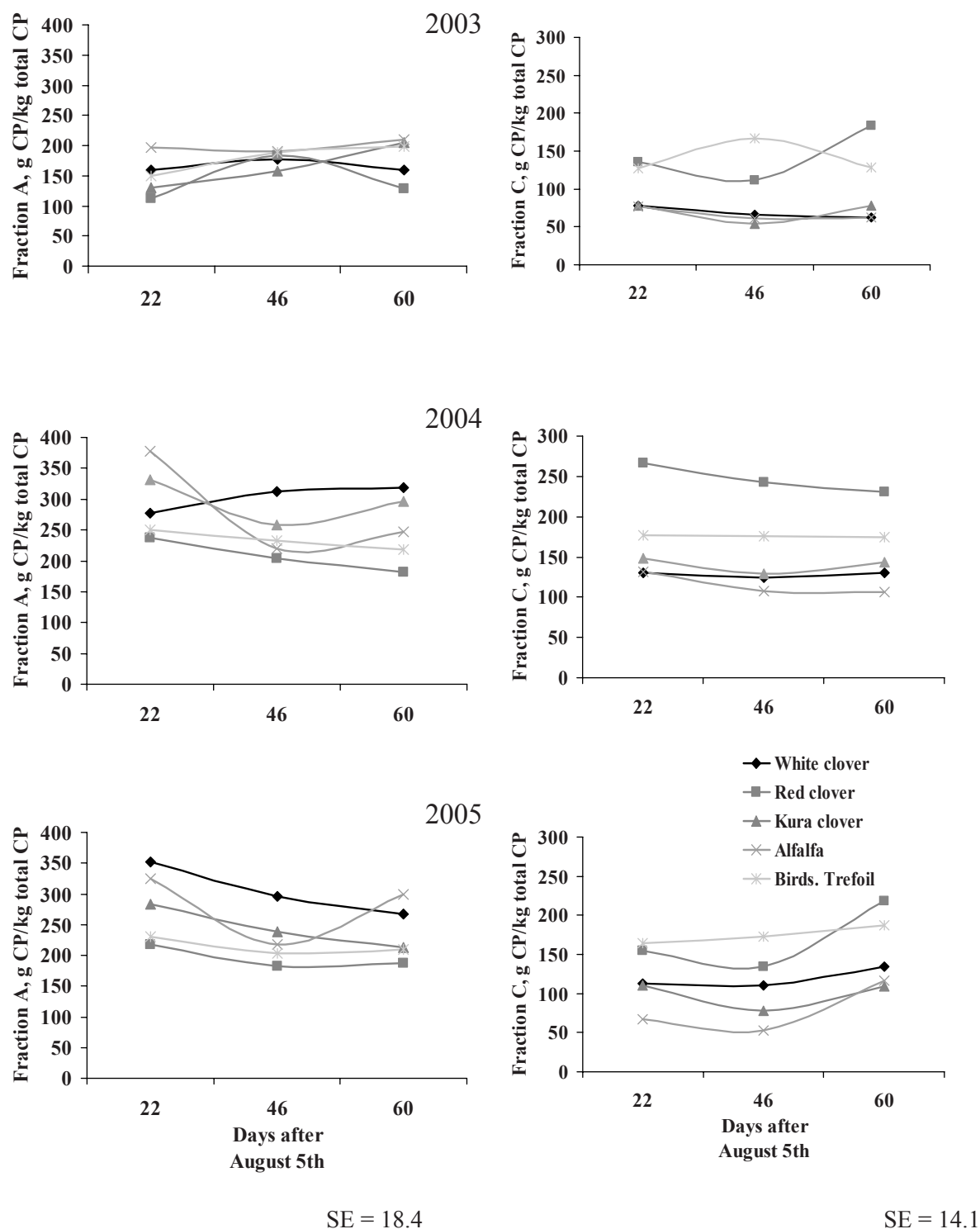


Figure 1. Content of fraction A (non protein N) and fraction C (in ADF residue bound N) of different legume species during the autumn growth in different years. SE is for Fraction A and C.



## Results and discussion

The botanical composition of the sward in the autumn growth showed a high proportion of legumes in the mixtures. The legume content declined with sampling date, but never fell below 40% (on a DM basis). The highest proportion was observed for red clover and alfalfa, with values ranging between 60 and 70%.

For the forage quality measurements, the analysis of variance revealed a significant effect of the interaction of species x year x sampling date. For CP, white clover and alfalfa showed the highest values, ranging between 20 and 29% for white clover and between 24 and 30% for alfalfa, confirming earlier observations (Stockdale, 1999). For all legume species, year effect showed generally similar values for 2003 and 2004, but higher values for 2005. The determination of cell wall components showed a large dependence of the year. In 2003 lower contents of NDF and ADF were observed, and higher values were observed in 2004 and 2005. White clover and kura clover had, in general, the lowest contents of cell wall components. The ADF contents were in the range 17-26% for white clover and 18-24% for kura clover. The NDF content varied between 32 and 45% for white clover, and 34 and 43% for kura clover. Although the interaction was significant, the NEL contents in all species were above 6 MJ kg<sup>-1</sup> DM. The age of legumes (days after August 5) did not show a marked effect. As shown in Figure 1, the fraction A was strongly dependent of the year effect. During autumn regrowth the course of fraction A development showed no clear trend for all legume species. Whereas alfalfa showed a curvilinear progress, other legumes species were decreasing or were invariable in the fraction A content. Red clover and birdsfoot trefoil showed, in general, the lowest contents of fraction A. White clover and alfalfa had fraction A contents in 2004 and 2005 above 300 g CP kg<sup>-1</sup> CP. The content of fraction C in forage legumes varied markedly between species and years. Red clover and birdsfoot trefoil showed, in general, the highest contents. White clover and kura clover had contents of fraction C that were sometimes below 100 g CP kg<sup>-1</sup> CP.

## Conclusion

The quality of legumes growing in autumn is largely dependent on the year, species and cutting date (in that order). Red clover and birdsfoot trefoil were shown to have more beneficial forage quality related to their crude protein fractionation, and higher N-use efficiency may be expected with these forage legume species in ruminant nutrition. In considering the protein quality, alternative legumes to white clover are therefore available. However, the suitability of each species for the defoliation systems of choice in autumn, e.g. grazing or cutting for silage, may be restricted by the low persistence of individual legume species.

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# Changes in water soluble carbohydrates for perennial ryegrass in the first growth in Norway and Sweden

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## Abstract

The objective of the paper was to determine the influence of seasonal changes in temperature and radiation on water soluble carbohydrates (WSC) in first growth of varieties of perennial ryegrass (*Lolium perenne* L.). This was done in small-plot field experiments with a three-cut silage system at two sites, one in Saerheim, Norway and one in Uppsala, Sweden. Perennial ryegrass varieties AberDart, which had been bred to accumulate high levels of WSC, and Fennema, which accumulates normal levels of WSC, were common to both sites. Trials were established in 2000 or 2001 and samples were taken during the two following years and every week three times before first harvest.

The results show that, on most sampling occasions during the first growth period at Uppsala, the WSC content of AberDart was significant higher than that of Fennema. In Saerheim, there were no differences between AberDart and Fennema during the two years, but the variety Tove had at some occasions a significant higher WSC content than AberDart. The average WSC levels varied between the sites, and the variations between years were larger in Saerheim than in Uppsala.

Keywords: *Lolium perenne*, water soluble carbohydrates (WSC), temperature, global radiation

## Introduction

Grasses with high levels of water soluble carbohydrates (WSC) have been shown to enhance livestock production (Miller *et al.*, 1999). This has led to the development of perennial ryegrass (*Lolium perenne* L.) cultivars that can accumulate high levels of WSC. The appeal of the high-WSC grasses lies in their potential to make protein synthesis in the rumen more efficient through matching protein and energy supply more closely, and so help reduce nitrogen losses to the environment. However, it is not known whether the genetic potential to accumulate high levels of WSC is expressed under different environments. The objective was to determine the influence of seasonal changes in temperature and radiation on WSC content in the first growth of different varieties of perennial ryegrass.

## Materials and methods

Two small-plot field experiments with an intended three-cut silage system were established in 2000 in Saerheim (58°47'N, 5°41'E, altitude 80 m), Norway and in 2001 in Uppsala (59°49'N, 17°39'E, altitude 5 m), Sweden. The experiments were of a single-factor randomized block design with four replicates and eight varieties, but only three are reported here for each site. At both sites, the diploid perennial ryegrass varieties AberDart, bred for high WSC accumulation, and the control variety Fennema were established. In addition to this, local varieties were established. The tetraploid varieties Tove in Saerheim and Helmer in Uppsala are reported here. Plots were fertilized in spring with 100 and 120 kg N ha<sup>-1</sup> in Uppsala and Saerheim respectively. P and K were applied according to the plant available levels in the soil.



Pre-harvest samples were taken by hand during first growth every week three times before first harvest to follow changes in the WSC content. All plots were harvested at 5 cm stubble height when Fennema had reached the early-boot stage using a Haldrup plot harvester (J. Haldrup, Lögstör, Denmark). In Uppsala, the first cut was taken 5 June 2002 (week 23) and 10 June 2003 (week 24). In Saerheim, first cut was taken 25 June 2001 (early week 26) and 3 June 2002 (week 23). All sampling and harvesting were done around 1400h and analysed for WSC content. Immediately upon cutting, 1 kg sub-samples were placed on ice and transported to the laboratory whereupon the samples were dried at 60 °C. Dried samples were milled through a 1 mm steel mesh and analysed for dry matter (DM) and WSC. NIRS (Near infrared spectroscopy) was used for determination of WSC with a common equation. The SAS procedure Mixed was used (SAS, 1997) for the statistical analysis of data with variety as fixed factor and replication as random factor.

## Results

Weather data for the periods of the investigation are presented in Table 1.

Table 1. Weather at the two experimental sites. Bold data indicate sampling period and one week before the first sampling.

Site and week number	First year			Second year		
	T <sub>mean</sub> (°C)	Precipitation (mm)	Sum global radiation (MJ m <sup>-2</sup> )	T <sub>mean</sub> (°C)	Precipitation (mm)	Sum global radiation (MJ m <sup>-2</sup> )
<b>Saerheim</b>						
18 (early May)	8.1	18.5	140	11.1	32.7	104
19 (mid May)	14.3	0.2	141	<b>18.7</b>	<b>16.0</b>	<b>121</b>
20 (mid May)	9.7	8.7	116	<b>12.1</b>	<b>4.4</b>	<b>115</b>
21 (late May)	<b>10.9</b>	<b>1.9</b>	<b>136</b>	<b>18.2</b>	<b>7.6</b>	<b>118</b>
22 (early June)	<b>10.3</b>	<b>11.1</b>	<b>116</b>	<b>14.1</b>	<b>13.1</b>	<b>150</b>
23 (early June)	<b>9.7</b>	<b>22.7</b>	<b>106</b>	<b>22.6</b>	<b>5.4</b>	<b>170</b>
24 (mid June)	<b>11.8</b>	<b>0.1</b>	<b>169</b>			
25 (late June)	<b>12.0</b>	<b>31.3</b>	<b>117</b>			
<b>Uppsala</b>						
18 (early May)	9.0	28.7	84	4.9	49.0	49
19 (mid May)	<b>11.2</b>	<b>0.0</b>	<b>176</b>	10.8	8.5	136
20 (mid May)	<b>11.8</b>	<b>10.6</b>	<b>142</b>	<b>10.6</b>	<b>2.5</b>	<b>139</b>
21 (late May)	<b>12.8</b>	<b>1.0</b>	<b>144</b>	<b>12.8</b>	<b>17.5</b>	<b>132</b>
22 (early June)	<b>14.4</b>	<b>0.0</b>	<b>171</b>	<b>13.7</b>	<b>6.0</b>	<b>151</b>
23 (early June)	<b>18.1</b>	<b>0.0</b>	<b>179</b>	<b>17.9</b>	<b>0.0</b>	<b>164</b>
24 (mid June)				<b>13.3</b>	<b>40.5</b>	<b>111</b>

T<sub>mean</sub> = weekly mean temperature.

Even though the sampling period was two weeks later at Saerheim in the first year, the temperature was lower than in the sampling period in the second year. In Saerheim the radiation sum was similar for both years. In Uppsala, temperature was similar for both years, but radiation sum was higher in the first year. The results in Table 2 for Uppsala show that on most sampling occasions during the first growth period, the WSC content of AberDart was significant higher than that of Fennema. In Saerheim, there were no differences between AberDart and Fennema during the two years, but the variety Tove had, on some occasions, a significantly higher WSC content than AberDart. The average WSC levels varied between the sites, and the variation between years was larger in Saerheim than in Uppsala. There was generally an increase in the WSC content from the start of the sampling until one week before

the first harvest, but during the week before the harvest the WSC content decreased in most situations.

Table 2. WSC ( $\text{g kg}^{-1}$  DM) at the two sites in first growth period.

Site and variety	First growth in the first year				First growth in the second year			
	3 weeks before FH	2 weeks before FH	1 week before FH	First harvest (FH)	3 weeks before FH	2 weeks before FH	1 week before FH	First harvest (FH)
<b>Saerheim</b>								
AberDart	298	321	321	305	149	209	208	199
Fennema	291	316	324	292	130	181	192	201
Tove	324	352	332	294	165	232	217	222
CV %	7.4	2.4	6.7	5.8	12.3	16.2	13.4	16.8
<i>P</i>	0.017	0.001	0.062	0.001	0.013	0.036	0.003	0.001
LSD	33	12	-	25	27	47	40	48
<b>Uppsala</b>								
AberDart	181	221	208	182	189	194	213	204
Fennema	162	192	184	139	165	157	176	160
Helmer	173	213	186	159	185	174	181	155
CV %	5.5	6.7	4.8	5.6	7.3	5.7	6.7	4.4
<i>P</i>	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
LSD	15	22	14	14	20	15	19	11

w = weeks, CV = coefficient of variation, LSD = least significant difference at  $P < 0.05$ .

## Discussion and conclusions

There were differences between the sites in the WSC relations between AberDart and Fennema. At Uppsala, AberDart had always a significantly higher WSC content than Fennema but, on the contrary in Saerheim, there were no differences between AberDart and Fennema during the two years. A much higher level in the WSC content in the first year in Saerheim could be explained by the lower temperature during the sampling period. A higher radiation level in first year in Uppsala has not influenced the WSC levels.

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## Endophytic fungi (*Neotyphodium*) in grasses. What is the situation in Sweden?

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### Abstract

Symbioses between grasses and endophytic fungi, *Neotyphodium* spp., have been studied only recently and to a small extent in Sweden. This paper summarizes the available data from the literature on grass endophytes in Swedish grasslands. The data comprise infection frequency of *Neotyphodium* in 10 species of grasses in natural sites and an agricultural site. Infection was screened with an immunoassay and microscopy. All five studied species of *Festuca* were infected. *Festuca pratensis* cv. Kasper had about 50% infection frequency and within individual infected plants the concentration of fungus increased during the growing season. In the single collection of *Phleum pratense* no infection was observed. The high infection frequency of *F. pratensis*, and the limited information on *Phleum pratense* as well as other important forage grasses, calls for further studies in Sweden. Effects of *Neotyphodium* on physiology, productivity and overwintering of cultivated grasses are so far not known.

### Introduction

Fungi in the genera *Neotyphodium* and *Epichloë* have become synonymous with 'fungal endophytes in cool season grasses'. *Neotyphodium* grows internally throughout the shoot, and is transmitted to a next grass generation via infected seeds, so-called vertically-transmitted asexual spreading. Increased plant vigour by *Neotyphodium* is reported for some grasses in warm climates. Most strikingly, *Neotyphodium* produces alkaloids with an anti-herbivore effect. Different grass species harbour different *Neotyphodium* species which produce alkaloids differing in their toxicity to insects and mammals. Several decades ago, livestock toxicoses in USA and New Zealand were first demonstrated to be attributable to alkaloids produced by *Neotyphodium* in introduced *Festuca arundinacea* and *Lolium perenne*. In Sweden, however, grass-endophyte symbioses have so far received only limited interest. The aim of this paper was to summarize the data reported so far for Swedish grasslands.

### Material and methods

As the subject is new in Sweden only four publications were available. Ten species from seven genera of grasses have been studied. Grass tillers collected in the field, or tillers of young plants raised from seeds collected in the field, have been analysed for presence of *Neotyphodium*. A commercially available immunoassay (Agrinostics, Watkinsville, GA, USA) was used. The antibodies in this immunoblot kit were raised against cell wall proteins from *Neotyphodium coenophialum* isolated from *Festuca arundinacea*, but the assay can be used also for other grass species (Koh *et al.*, 2006). In the Swedish studies microscopy of grass tillers has been used for comparison or to confirm results from immunoblots. The immunoassay was also used quantitatively to determine the concentration of *Neotyphodium uncinatum* in *Festuca pratensis* by calibrating the intensity of imprints in the immunoblots against counted number of hyphae across grass leaf sheaths.

## Results and discussion

Among grasses collected from non-agricultural sites (Table 1) there was no infection in *Agrostis capillaris* and no, or very low, infection in *Deschampsia flexuosa* and *Poa trivialis*. On the other hand, the *Festuca* species *F. ovina*, *F. rubra* and *F. vivipara* showed infection frequencies of 10-37%. In *Festuca rubra* infection frequency decreased significantly at higher altitudes (Bazely *et al.*, 2007).

Table 1. *Neotyphodium* infection frequency of native and agricultural grasses in Sweden. Infection was determined by immunoassay (I) or microscopy (M).

Grass species, cultivar	Site	Infection frequency (%)	No. of samples	Reference
<i>Agrostis capillaris</i>	Non-agricultural			
	All over Sweden, 57 sites	0.5 (I); 0 (M)	209; 135	Bazely <i>et al.</i> 2007
<i>Deschampsia flexuosa</i>	All over Sweden, 57 sites	4 (I); 0 (M)	71; 165	Bazely <i>et al.</i> 2007
<i>Festuca ovina</i>	Abisko	13 (I)	1598	Granath <i>et al.</i> 2007
	Tärnaby-Sälen	15 (I)	1268	
<i>Festuca rubra</i>	All over Sweden, 57 sites	27 (I); 23 (M)	312; 342	Bazely <i>et al.</i> 2007
<i>Festuca rubra</i>	Abisko	10 (I)	799	Granath <i>et al.</i> 2007
	Tärnaby-Sälen	23 (I)	1204	
<i>Festuca vivipara</i>	Abisko	12 (I)	470	Granath <i>et al.</i> 2007
	Tärnaby-Sälen	37 (I)	107	
<i>Poa trivialis</i>	All over Sweden, 57 sites	4 (I); 0 (M)	241; 154	Bazely <i>et al.</i> 2007
Agricultural				
<i>Dactylis glomerata</i> cv. Laika	Umeå	37.5 (I); 0 (M)	26	Koh <i>et al.</i> 2006
<i>Festuca ovina</i>	Umeå	23 (I); 23 (M)	13	Koh <i>et al.</i> 2006
<i>Festuca pratensis</i>	Umeå	50 (I); 50 (M)	20	Koh <i>et al.</i> 2006
<i>Festuca pratensis</i> cv. Kasper	Umeå	25-65 (I)	162	Puentes <i>et al.</i> 2007
<i>Phalaris arundinacea</i> cv. RF9402	Umeå	60 (I); 0 (M)	10	Koh <i>et al.</i> 2006
<i>Phleum pratense</i> cv. Jonatan	Umeå	0 (I); 0 (M)	39	Koh <i>et al.</i> 2006

Five species were collected from agricultural experimental fields (Table 1) in Umeå (63.75°N, 20.28°E). *Phleum pratense* did not show any infection. *Dactylis glomerata* and *Phalaris arundinacea* gave inconclusive results when immunology and microscopy were compared. *Festuca pratensis* cv. Kasper was highly infected, about every second plant contained *Neotyphodium*. *Festuca ovina* is the only species that was collected from both natural sites and from agricultural soil. This species was infected in both types of sites.

The frequent infection of the genus *Festuca*, both in nature and in agriculture, is evident in the Swedish collections and in a Finnish survey (Saikkonen *et al.*, 2000). Infection frequencies in *Festuca ovina*, *F. rubra* and *F. pratensis* are in good agreement in the Swedish and Finnish studies. In the Finnish study *Deschampsia flexuosa* had < 20% infection and *Phalaris arundinacea* had no infection. In contrast, *Phleum pratense* showed an infection frequency of 33% in natural populations in Finland (Saikkonen *et al.*, 2000), but no infection in the Swedish plants (Table 1). This calls for further examination of the very important forage grass *Phleum pratense* in Sweden. Another difference between the two countries was the high (67%) infection frequency in *Agrostis capillaris* in Finland but no infection in Sweden. High variation among populations and few sampled populations are likely explanations.

In a Finnish study, two seed lots of *Festuca pratensis* cv. Kasper had 41% and 92% infection frequency while other varieties of this grass showed either close to 100% or almost no infection (Saikkonen *et al.*, 2000). Also, when several varieties of *F. pratensis* were sampled in Umeå there was a large variation of infection frequency (Puentes *et al.*, 2007). It was 0 in cv. Hibinskaja, 30% in cv. Kasper, 60% in cv. Norild and 90% in cv. Inkeri. However, small sample numbers and the poorly known history of seed storage conditions prevented any firm conclusions, especially when infection frequency was low. Storage of seeds for a long time as well as moist or warm storage can reduce viability of *Neotyphodium* in seeds. Methods for detection of *Neotyphodium*, i.e. immunoassays, microscopy and DNA-based techniques, are time-consuming and can not distinguish between viable and non-viable fungi. To determine if fungi are viable seeds need to be germinated and the seedlings then examined for presence of *Neotyphodium*.

Seasonal variation in fungal concentration was studied in individual plants by sampling tillers from the same infected *F. pratensis* plants repeatedly over a growing season (Puentes *et al.*, 2007). The concentration of *Neotyphodium* in each plant increased about 3-fold during the growing season. The physiology of the *F. pratensis*  $\times$  *Neotyphodium* symbiosis is very poorly known and it is presently difficult to explain variations in endophyte concentration. However, it is worth mentioning that the increase in endophyte concentration was significantly correlated to cumulative degree days during the growing season.

When considering that large agricultural areas are used for grasslands in Sweden it is surprising that grass endophytes have been largely neglected until recently. *Neotyphodium* does not give any visible symptoms on the grass, and no serious problem with livestock which were clearly due to *Neotyphodium* has been reported (P. Häggblom, National Veterinary Institute, Uppsala, pers. comm.). Therefore, occurrence of grass endophytes may have been overlooked. The alkaloid loline, which is produced by *Neotyphodium uncinatum* in *F. pratensis*, is poisonous to insects but not to large mammals (Schardl and Panaccione, 2005). Still, it may be that presence of *Neotyphodium* gives more subtle effects, e.g. decreased palatability which can lead to decreased forage consumption and reduced animal production.

## Conclusions

Endophytic fungi in grasses were clearly demonstrated in Sweden. In particular, *Festuca* species were infected by *Neotyphodium*. The high infection frequency in the forage grass *F. pratensis* calls for further studies of this species. The results of the single study of *Phleum pratense* differed from results in Finland, and this highly important forage species should be examined further. Studies of any effects of *Neotyphodium* on physiology, productivity and overwintering of cultivated grasses will be rewarding.

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## Quality of timothy (*Phleum pratense* L.) and causality of its variation

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### Abstract

Timothy (*Phleum pratense* L.) is high yielding, well adapted to northern European conditions, and is one of the most cultivated grass species in Lithuania. The concentrations of crude protein (CP), neutral detergent fibre (NDF), water soluble carbohydrates (WSC), and dry matter digestibility (DMD) in herbage of *P. pratense* at early heading stage varied in relation to the weather conditions of the growing year. The CP concentration varied within a range of 114-145, NDF 574-596, WSC 54-139 and DMD 510-575 g kg<sup>-1</sup>. Plant maturity is the factor that exerts the greatest effect on quality. The onset of maturity during the early growth stage in spring was accompanied by significant changes in nutritive value: increase in NDF, declines in CP and DMD. The dynamics of quality changes depends on the earliness of varieties. Plants were found to differ in quality parameters of separate plant parts: stems contained more fibre, WSC, less CP and DMD than leaves.

Keywords: *Phleum pratense*, quality parameters, stems, leaves

### Introduction

Due to adaptation to the cool and relatively humid northern climate, timothy (*Phleum pratense*) is an important forage grass that is widely grown in the regions with changeable and severe over wintering conditions, including Lithuania, where it is one of the most productive grasses at the first cut. Timothy is valued for its very high winter hardiness, tolerance of waterlogged soil and good palatability. The nutritive value of timothy is affected by genotype (Bregard *et al.*, 2001), maturity stage (Jefferson, 2004), fertilization (Belanger *et al.*, 2004), morphological peculiarities (Cleassens *et al.*, 2005), and climatic conditions (Bertrand *et al.*, 2006). Lithuanian-bred timothy varieties and breeding lines differ in earliness, herbage and seed yield, leafiness and other traits. The aim of this work was to study the causality of variation of quality parameters of Lithuanian-origin timothy at different maturity stages and various plant parts in different growing years.

### Materials and methods

Quality testing experiments were assessed over the period 2003-2007 (except 2005). Altogether 6 varieties and 51 breeding lines of timothy (of Lithuanian origin) were tested. Quality testing trials were established annually and were evaluated at the spring – summer growth cycle in the first year of herbage utilization. The experiments were established on carbonate gleyic, moderately heavy drained brown soil. The plough layer was 25-30 cm, and soil had the following properties: pH 7.2-7.5, humus 1.9-2.2%, total nitrogen 0.14-0.16%, mobile phosphorus 201-270 mg kg<sup>-1</sup>, and potassium 101-175 mg kg<sup>-1</sup> of soil. Samples of 51 breeding lines were taken once, at early heading of each genotype. Samples of the 6 varieties were taken 3 times: the first time they were taken simultaneously at booting of the plants, and the second and third times they were taken for each genotype individually at early heading and full heading. Samples for chemical analyses were dried and ground by a mill with 1 mm sieve, and analysed by NIR spectroscopy. The concentrations of crude protein (CP), neutral detergent fibre (NDF), water soluble carbohydrates (WSC), and dry matter digestibility (DMD) were determined. The statistics of equations for the prediction of quality components

(coefficients of correlation in calibration RSQ and standard errors in calibration SEC, g kg<sup>-1</sup>) are: 0.95 and 8.6 for CP, 0.98 and 13.8 for NDF, 0.97 and 12.2 for WSC, 0.93 and 27.1 DMD, respectively. Structural analyses of plant parts were performed at grass full-heading stage of the first cut.

## Results and discussion

The data of herbage nutritive quality parameters of *P. pratense* at the early heading over the period 2003-2007 (except 2005) and coefficient of variation (CV %) are presented in Table 1.

Table 1. Mean (g kg<sup>-1</sup>) and coefficient of variation of quality characteristics of timothy at the early heading.

Variable	2003		2004		2006		2007		2003-2007	
	mean	CV%	mean	CV%	Mean	CV%	mean	CV%	mean	CV%
CP	123	6.39	114	8.26	127	8.01	145	7.45	126	11.71
NDF	574	3.54	580	4.56	577	3.91	596	2.38	581	3.96
WSC	137	14.91	139	13.63	139	13.41	54	30.40	120	32.62
DMD	575	7.27	571	6.25	553	5.31	510	6.03	552	7.40

During the experimental period the herbage quality varied inappreciably: crude protein (CP) fluctuated within 114-145 g kg<sup>-1</sup>, neutral detergent fibre (NDF) within 574-596 g kg<sup>-1</sup>, and dry matter digestibility (DMD) within 510-575 g kg<sup>-1</sup>. Water soluble carbohydrates (WSC) concentration was found to be most variable parameter among the herbage quality parameters tested: subject to growing year variation coefficient was as high as 13.41-30.40%. Mean values and range of variation could have been affected by each year's different growing conditions. WSC levels are usually the highest in plants grown in high light intensity and low temperatures (Bertrand *et al.*, 2006). The weather during the growing season in 2003, 2004 and 2006 was sunny and cool which resulted in WSC values in the range 101-173 g kg<sup>-1</sup>, whereas in 2007, when there was less sunshine and higher air temperatures, WSC declined to 37-72 g kg<sup>-1</sup>. The nutritive value varies depending on plant growth stage (Jefferson, 2004). Quality parameters of timothy were assessed at different plant maturity stages (Table 2).

Table 2. Variation of quality characteristics of timothy at different maturity stages.

Variable	Maturity stage								
	Booting			Early heading			Full heading		
	Mean, g kg <sup>-1</sup>	Range, g kg <sup>-1</sup>	CV%	Mean, g kg <sup>-1</sup>	Range, g kg <sup>-1</sup>	CV%	Mean, g kg <sup>-1</sup>	Range, g kg <sup>-1</sup>	CV%
CP	167	150-176	7.40	117	99-138	9.70	96	84-106	8.06
NDF	540	512-571	4.05	643	594-669	4.37	667	627-701	3.52
WSC	149	136-173	9.72	110	67-179	41.91	107	41-191	54.56
DMD	684	649-718	3.57	539	466-574	7.31	464	387-538	11.26

The data for CP, averaged over the period from early heading to full heading, indicate that it declined by 3.56 g kg<sup>-1</sup> per day. WSC content is generally higher in young grass but the coefficient of variation was low at this stage, while at more advanced growth stages WSC varies within a wider range. NDF increased from 540 to 667 g kg<sup>-1</sup>, whereas DMD decreased from 684 to 464 g kg<sup>-1</sup> from booting to full heading. The variation of quality indicators (except WSC) at booting and early heading was minor. Many authors suggest that herbage quality parameters correlate strongly with plant structure composition, especially leafiness (Tarakanovas *et al.*, 2006). Stem growth is important for yield increase in timothy, but a high amount of stems causes low DMD. We estimated quality parameters of different plant parts – leaves, stems and inflorescences (Table 3).



Table 3. Variability of quality parameters of plant parts.

Variable	Leaves			Stems			Inflorescences		
	Mean, g kg <sup>-1</sup>	Range, g kg <sup>-1</sup>	CV%	Mean, g kg <sup>-1</sup>	Range, g kg <sup>-1</sup>	CV%	Mean, g kg <sup>-1</sup>	Range, g kg <sup>-1</sup>	CV%
CP	148	136-160	5.31	40	32-46	10.79	107	98-116	6.03
NDF	596	571-631	3.57	753	703-799	4.07	629	603-675	3.69
WSC	32	17-61	51.26	104	83-134	17.99	38	25-63	37.17
DMD	519	494-541	2.87	390	324-432	10.71	372	352-384	3.63

The highest level of, CP 148 g kg<sup>-1</sup>, was identified in leaves. There was 28% (calculated as 148-107/148) less CP in inflorescence than in leaves and 73% (148-40/148) less CP in stems than in leaves. The highest levels of NDF and WSC, 753 and 104 g kg<sup>-1</sup>, were determined in stems. Coefficient of variation of DMD, CP and NDF of stems was higher than in leaves or inflorescences. WSC concentration was the most variable quality indicator in leaves, ranging from 17 to 61 g kg<sup>-1</sup>, whereas DMD varied the most in stems.

### Conclusion

A great amount of variation in WSC concentration was found in *Phleum pratense* varieties and breeding lines over the experimental period. Variation of quality parameters of timothy at different plant maturity stages was not high except for WSC. NDF and DMD varied the most in stems. Based on these results, leafiness and high stem quality should be the main criteria for selecting accessions for timothy breeding. On the other hand, a proper choice of cutting regime for the individual variety may be the key for practical management of timothy in terms of matching good quality and yield.

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# Contents of phenolics, protein precipitation capacity of tannin in browse and ability to predict them in diets from faecal composition in sheep

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## Abstract

The potential of faecal composition to predict the content and biological effects of tannin in *Acacia saligna*, *Chamaecytisus palmensis* and *Atriplex amnicola* diets were investigated in 3 feeding trials. Each of the 19 experimental diets (browse and *Avena sativa*) was fed to 6 replicate sheep. Total phenolics (TPC) and tannin (TTC) contents, protein precipitation capacity of tannin (PPC), *in-vitro* gas production (*ivGP*), *in-vitro* organic matter digestibility (*ivOMD*) and *in-vitro* metabolizable energy content (*ivME*) of the experimental diets were determined. Respective faeces were analysed for proximate composition. Data from the 3 trials were pooled to generate predictive models. The TTC of *A. saligna* was greater (36.2 g kg<sup>-1</sup> dry matter (DM)) than *C. palmensis* and *A. amnicola* (6.4 g kg<sup>-1</sup> DM and 1.1 g kg<sup>-1</sup> DM). Tannin of *A. saligna* reported the highest PPC (27.3 g kg<sup>-1</sup> DM) and thereby reduced the *ivGP*, *ivOMD* and *ivME*, significantly ( $P < 0.05$ ). Biological effects of tannin in *C. palmensis* and *A. amnicola* were not significant ( $P < 0.05$ ). Strong linear relationships ( $P < 0.0001$ ) were found between faecal acid detergent fibre, acid detergent lignin and N contents with TPC, TTC and PPC of the diets. Anti-nutritive effects of tannin and the ability of faecal composition to predict phenolics contents and PPC of browse diets were evident in sheep.

Keywords: browse, tannin, biological effects, faecal predictions

## Introduction

Phenolics, particularly the tannins that occur in forage browse species, form insoluble complexes with protein and fibre, thus they remain undegraded by rumen flora limiting their availability to ruminants. In tropical browse species, total phenolics (TPC), total tannin (TTC) and condensed tannin (CT) contents range from 17-250, 7-214 and 0-260 g kg<sup>-1</sup> dry matter (DM), respectively, while protein precipitation capacity of tannin (PPC) ranges from 0-0.0011 g kg<sup>-1</sup> DM (Getachew *et al.*, 2002). *Acacia saligna* is high in TPC, TTC and CT (Degen *et al.*, 2000), while *Chamaecytisus palmensis* is low in tannin (Kaitho *et al.*, 1998). Very low tannin content is reported in *Atriplex amnicola* (Norman *et al.*, 2004). Phenolics in browse species increase towards summer and decrease towards winter (Edwards, 2000; Salem, 2005). A significant proportion of CT is bound with fibre fractions in *A. saligna* leaves and in the faeces of sheep (Makkar *et al.*, 1995). Free CT in *A. saligna* get bound to fibre and protein in the ruminant digestive tract (Degen *et al.*, 2000). Tannins of *C. palmensis* (Kaitho *et al.*, 1998) and *A. saligna* (Krebs *et al.*, 2007) enhance the excretion of faecal N in sheep. The objective of this study was to test the hypothesis that faecal composition has the potential to predict contents and protein precipitation capacity of tannin in browse diets fed to sheep.

## Materials and methods

In 3 pen-feeding trials, 6 DM levels of *A. saligna* (0, 250, 480, 690, 840, 1000 g kg<sup>-1</sup>), 7 DM levels of *C. palmensis* (0, 180, 350, 520, 720, 870, 1000 g kg<sup>-1</sup>) and 6 DM levels of *A. amnicola* (0, 160, 360, 500, 650, 840 g kg<sup>-1</sup>) were fed to sheep. *Avena sativa* (oaten) hay

was used to balance the DM contents of the diets. Each diet was fed to 6 replicate sheep for 10 days; 7 days for adaptation followed by 3 days collection period. Freeze-dried, composite feed samples were analysed. The TPC and TTC were determined by folin-ciocalteu method (Makkar, 2003). The PPC was determined using bovine serum albumin, and *in-vitro* gas production (*iv*GP) was measured both in the presence and in the absence of polyethylene glycol (PEG) in tannin bioassay (Makkar, 2003). *In-vitro* metabolizable energy (*iv*ME) and organic matter digestibility (*iv*OMD) were calculated from *iv*GP (Menke and Steingass, 1988). Composite oven-dried faecal samples were analysed for proximate composition. Significance of biological effects of tannins was tested by comparing *iv*GP, *iv*ME and *iv*OMD in the presence and absence of PEG using *t* test. Predictive regression models were fitted using pooled data and the best fit models were selected considering F statistics,  $R^2$ , standard deviation of error and *t* statistics of estimates in the ANOVA procedure.

## Results and discussion

Having a greater amount of tannin, *A. saligna* showed a greater PPC than both *C. palmensis* and *A. amnicola* (Table 1). Nevertheless, TPC, TTC and PPC of *A. saligna* shown in this study were lower than previous reports (Degen *et al.*, 2000; George *et al.*, 2007). This result might be due to conducting the trial with *A. saligna* during late autumn-mid winter period (Salem, 2005) and morphological variation of *A. saligna* (George *et al.*, 2007).

Table 1: Chemical composition and biological effects of tannin of forage species.

Parameter		<i>Acacia saligna</i> <sup>†</sup>	<i>Chamaecytisus palmensis</i>	<i>Atriplex amnicola</i>	<i>Avena sativa</i>
NDF (g kg <sup>-1</sup> DM)		598.8	631.8	516.3	619.7
CP (g kg <sup>-1</sup> DM)		112.7	130.3	129.9	51.6
TPC (g kg <sup>-1</sup> DM)		46.5	24.2	3.0	9.9
TTC (g kg <sup>-1</sup> DM)		36.2	6.4	1.1	2.5
PPC (g kg <sup>-1</sup> DM)		27.3	0.1	1.0	0.4
<i>iv</i> GP (ml 200mg <sup>-1</sup> DM)	PEG –	17.8	43.2	24.3	45.2
	PEG +	29.3	40.7	25.8	44.8
<i>iv</i> ME (MJ kg <sup>-1</sup> DM)	PEG –	5.3	8.8	6.3	8.6
	PEG +	6.8	8.5	6.5	8.6
<i>iv</i> OMD(MJ kg <sup>-1</sup> DM)	PEG –	361.3	594.9	437.4	576.4
	PEG +	463.8	571.6	449.6	573.7

TPC, total phenolics; TTC, total tannin; PPC, protein precipitation capacity of tannin; NDF, neutral detergent fibre; CP, crude protein; DM, dry matter; *iv*GP, in-vitro gas production; *iv*ME, in-vitro metabolizable energy; *iv*OMD, in-vitro organic matter digestibility; PEG+, with polyethylene glycol; PEG–, without polyethylene glycol, †: biological effect of tannin was significant ( $P < 0.05$ ).

Table 2: Predictive regression models ( $Y_i = a + b X_i$ ) of total phenolics, and tannin contents and protein precipitation capacity of tannin ( $P < 0.0001$ ).

$Y_i$	$X_i$	$a \pm SE$	$b \pm SE$	Standard deviation of error	Coefficient of variation	$R^2$
TPC	<i>fc</i> ADF	$-57.25 \pm 6.39$	$0.15 \pm 0.01$	6.87	26.36	0.79
TPC	<i>fc</i> ADL	$-2.48 \pm 2.53$	$0.12 \pm 0.01$	7.23	27.73	0.76
TPC	<i>fc</i> N	$-18.28 \pm 3.54$	$2.65 \pm 0.20$	6.93	26.58	0.78
TTC	<i>fc</i> ADF	$-61.64 \pm 6.18$	$0.14 \pm 0.01$	6.64	37.79	0.78
TTC	<i>fc</i> ADL	$-10.08 \pm 2.29$	$0.12 \pm 0.01$	6.54	37.22	0.79
TTC	<i>fc</i> N	$-24.66 \pm 3.40$	$2.53 \pm 0.20$	6.65	37.87	0.78
PPC	<i>fc</i> ADF	$-51.59 \pm 5.85$	$0.11 \pm 0.01$	6.23	62.03	0.71
PPC	<i>fc</i> ADL	$-11.78 \pm 2.08$	$0.09 \pm 0.01$	5.95	58.69	0.74
PPC	<i>fc</i> N	$-23.09 \pm 3.14$	$1.99 \pm 0.18$	6.15	60.64	0.72

TPC, total phenolics; TTC, total tannin; PPC, protein precipitation capacity of tannin; *fc*ADF, faecal acid detergent fibre; *fc*ADL, faecal acid detergent lignin; *fc*N, faecal N, contents (g kg<sup>-1</sup> dry matter).

The trials with *C. palmensis* and *A. amnicola* were conducted during the late winter-spring period; thus the results confirmed that the low levels of phenolics are characteristics of *C. palmensis* (Edwards, 2000) and *A. amnicola* (Norman *et al.*, 2004), particularly in the spring. As shown in Table 1, despite having greater contents of fibre and crude protein, *A. saligna* showed the lowest *iv*GP, *iv*ME and *iv*OMD, in the absence of PEG. However, out of the forage species studied only *A. saligna* showed a significant ( $P < 0.05$ ) increase of *iv*GP, *iv*ME and *iv*OMD in the presence of PEG. The TTC and PPC were much higher in *A. saligna*. Therefore, anti-nutritive biological effects were evident in the high tannin-containing *A. saligna*, but negligible in low tannin-containing *C. palmensis*, *A. amnicola* and *A. sativa*.

Consistent with the findings of Getachew *et al.* (2002), PPC was significantly correlated ( $P < 0.0001$ ;  $r > 0.91$ ) with TPC and TTC. Phenolics properties (TPC, TTC, PPC) of the diets were highly correlated ( $P < 0.0001$ ,  $r > 0.84$ ) with *fc*ADF, *fc*ADL and *fc*N contents. However, faecal neutral detergent fibre (*fc*NDF) and ash contents were less correlated ( $r < 0.77$ ) with the phenolics properties of the diets. Tannin in *A. saligna* binds with fibre in forage; thereby it will be present with fibre in faeces (Makkar *et al.*, 1995). Tannins decrease crude protein digestibility and increase the faecal N (Kaitho *et al.*, 1998; Krebs *et al.*, 2007). Significant linear regressions ( $P < 0.0001$ ) were found between *fc*ADF, *fc*ADL and *fc*N contents with TPC, TTC and PPC of experimental diets (Table 2). According to the standard deviation of error and  $R^2$  values, all predictive models were equally strong. Nevertheless, based on the coefficient of variation, it can be suggested that the robustness of predictions are in the decreasing order for TPC > TTC > PPC. The ability of predicting the TPC, TTC and PPC from faecal ADF, ADL and N contents were evident.

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# Improvement of quality of silage made of bird's foot trefoil by using additives and bird's foot-timothy mixture

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## Abstract

Bird's foot trefoil (*Lotus corniculatus*) is a leguminous forage. Legumes have a high nutritive value but they are known to be difficult to ensile, frequently resulting in poorly fermented silage. However, the use of the efficient additive may considerably improve the quality of silage and reduce losses. It is also possible to improve the fermentation properties through the use of grass-legume mixtures rather than pure legume. In this ensiling study herbage of pure bird's foot trefoil and of bird's foot trefoil-timothy mixture were ensiled without additives and also with the use of chemical additives AIV-2000 plus and KS-1 (5 l ton<sup>-1</sup> fresh matter). The results indicated that the nutritive value of the silage was dependent on the mixture. The pure bird's foot trefoil resulted in higher crude protein and metabolizable energy concentrations. The silage fermentation quality differed between the treatments with and without additives, and between the pure legume and the grass-legume mixture. The silage had the lowest quality when no additives were used and when pure bird's foot trefoil was ensiled.

Keywords: bird's foot trefoil, quality of silage, additive, mixture

## Introduction

Bird's foot trefoil (*Lotus corniculatus*) is a legume used for grazing and making silage. Forage legumes have a high nutritive value but they are known to be difficult to ensile and result often in poorly fermented silage. This is usually due to the high buffering capacity (BC), the low available sugar (WSC) and dry matter (DM) concentrations. Results have shown that the silage quality made of legume crops can be improved considerably by using an effective additive and/or replacing pure legume with a legume-grass mixture (Lättemäe *et al.*, 2000; Lättemäe and Meripõld 2005; Lättemäe *et al.*, 2006). There are also interaction effects when using additives with grass-legume mixtures. The aim of this investigation was to study the ensiling of bird's foot trefoil, the effects of an additive and legume-grass mixture effects on nutritive value, fermentation quality and dry matter losses of silage.

## Materials and methods

The ensiling trial was carried out at the Department of Grassland of ERIA on 20 June 2006. The first cut of a crop of bird's foot trefoil (variety Norcen) and of a bird's foot trefoil-timothy (variety Tika) mixture were used as the materials for the silage. The botanical composition the first treatment consisted of 85% bird's foot trefoil, with 9% weeds, 4% grasses and 2% pink clover. The mixture consisted of 50% of timothy, 49% bird's foot trefoil and 1% of weeds. The bird's foot trefoil was flowering at development stage and timothy at heading stage when harvested. The chemical composition of the bird's foot trefoil was as follows: dry matter concentration (DM) 211 g kg<sup>-1</sup>, crude protein (CP) 182 g kg<sup>-1</sup> DM, crude fibre (CF) 243 g kg<sup>-1</sup> DM, neutral detergent fibre (NDF) 381 g kg<sup>-1</sup> DM, acid detergent fibre (ADF) 294 g kg<sup>-1</sup> DM, water soluble carbohydrates (WSC) 59.2 g kg<sup>-1</sup> DM, buffering capacity (BC) 62.4 g lactic acid kg<sup>-1</sup> DM. The bird's foot trefoil-timothy mixture was DM 252 g kg<sup>-1</sup>, CP 122 g kg<sup>-1</sup> DM,



CF 283 g kg<sup>-1</sup> DM, NDF 510 g kg<sup>-1</sup> DM, ADF 334 g kg<sup>-1</sup> DM, WSC 76.0 g kg<sup>-1</sup> DM, and BC 58.9 g lactic acid kg<sup>-1</sup> DM.

The grass was cut by a mowing machine, chopped at the 2-3 cm chopping length and ensiled in 3-litre glass jars. Two different chemical additives KS-1 and AIV-2000 plus were used at an application rate of 5 l ton<sup>-1</sup> fresh matter. The control was the untreated variant. KS-1 is a new additive based on potassium sorbate and contains also sodium nitrite. AIV-2000 plus is based on formic acid. The trial was performed in three replicates. The jars were covered with plastic film (4 layers), and kept at room temperature of 18-25 °C and the samples were taken after 120 days.

The volatile fatty acids (VFA), ethanol, pH and ammonia N of the silage were determined from water solution. Ammonia N was determined by using Kjeldrec Autosystem 1030. Ethanol, VFA and butanediol were determined by using gas chromatograph (Faithfull, 2002). The concentrations of DM, CP and CF were determined from dried and ground samples (AOAC, 1990). WSC were determined by Bertran method (Thomas, 1977). NDF and ADF were determined by using ANKOM 200 test (Van Soest *et al.*, 1977). The DM losses were calculated from the differences of weight of the silage mass during fermentation. The aerobic stability of the silage was estimated visually at the room temperature of 25 °C. The recording time was taken as a base when the first colonies of mould appeared on the surface of silage.

## Results and discussion

The results are presented in the Tables 1 and 2. The nutritive value and the concentration of DM of silage were dependent on the material for ensiling. The pure bird's foot trefoil resulted in lower dry matter concentration and higher nutritive value. The average concentration of DM of the bird's foot trefoil was 204 g kg<sup>-1</sup>, CP 190 g kg<sup>-1</sup> DM and ME 9.9 MJ kg<sup>-1</sup> DM. In the mixture the concentration of DM slightly increased but the nutritive value decreased. There were large differences in the yields of DM (bird's foot trefoil 1,445 kg ha<sup>-1</sup> vs. mixture 3,769 kg ha<sup>-1</sup>).

The fermentation quality is usually dependent on the ensilability of the material. Considering the concentrations of DM and WSC of the fresh bird's foot trefoil and the buffering capacity, such material has moderate ensilability. In the mixture the properties of the fermentation slightly improved. Generally, the quality of silage was satisfactory or even good but there were significant differences between treatments. The fermentation quality was mainly dependent on the additive but also on the mixture. The untreated bird's foot trefoil silage had the lowest quality. It contained 2.3 g kg<sup>-1</sup> DM of butyric acid and 7.9% of ammonia. Both of the additives improved fermentation and reduced DM losses. The other treatments of the mixture had also better quality.

Table 1. Effects of additive and mixture on nutritive value, dry matter losses and aerobic stability of silage.

Treatment	DM g kg <sup>-1</sup>	CP g kg <sup>-1</sup> DM	CF g kg <sup>-1</sup> DM	ME g kg <sup>-1</sup> DM	DM losses %	Aerobic stability, days
<u>Bird's foot trefoil</u>						
Untreated	203	188	265	9.8	4.6	> 7
AIV-2000 plus	207	190	251	9.9	1.6	4.0
KS-1	203	191	256	9.9	1.7	> 7
<u>Bird's foot trefoil-timothy mixture</u>						
Untreated	234	123	298	9.6	4.5	4.0
AIV-2000 plus	243	125	286	9.5	1.4	4.5
KS-1	244	120	288	9.5	1.3	7
<i>LSD</i> <sub>0.05</sub>	26.6	21.1	16.5	0.3	2.3	

Table 2. Effects of additive and mixture on fermentation quality of silage.

Treatment	pH	Amn. N % total N	Acetic acid g kg <sup>-1</sup> DM	Butyric acid g kg <sup>-1</sup> DM	Ethanol g kg <sup>-1</sup> DM	Butanediol g kg <sup>-1</sup> DM
<u>Bird's foot trefoil</u>						
Untreated	5.5	7.9	16.8	2.3	35.0	0.8
AIV-2000 plus	5.2	7.4	10.4	1.0	34.0	0.0
KS-1	4.7	3.7	16.2	0.7	58.0	0.0
<u>Bird's foot trefoil-timothy mixture</u>						
Untreated	5.4	5.0	12.8	0.9	43.2	1.7
AIV-2000 plus	4.9	5.3	9.5	0.6	20.1	0.2
KS-1	4.7	4.3	15.4	0.4	43.3	0.0
<i>LSD<sub>0,05</sub></i>	<i>0.4</i>	<i>3.4</i>	<i>4.0</i>	<i>1.2</i>	<i>16.6</i>	<i>0.8</i>

The other fermentation products were in accordance with quality of silage. Ethanol is indicating the development of yeasts, butanediol the growth of enterobacteria and the acetic acid the trend of lactic acid fermentation. The DM losses were also in accordance with quality of silage. The aerobic stability of silage was dependent on the treatment. A good stability was obtained (7, > 7 days) when the additive KS-1 was applied to the silage. In other treatments the stability varied from 4 to 4.5 days.

## Conclusions

The results demonstrated that bird's foot trefoil is a legume that is of high nutritive value with properties that are moderately good for ensiling. In order to increase the DM yield of the harvested crop and its ensilability, it is recommended that it be grown with grasses. The use of both the additive, and of the timothy grass-bird's foot trefoil mixture relative to the pure legume, improved the fermentation and reduced DM losses. There was also a better aerobic stability when KS-1 was used.

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# Nutritive value in leaves and stems of lucerne with advanced maturity and a comparison of methods for determination of lignin content

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## Abstract

Contents of crude protein and cell-wall constituents in leaves and stems of lucerne (*Medicago sativa* L.) cv K-28 in three stages of growth were investigated. Sampling was done at 7-day intervals in the second regrowth. During growth and development, crude protein content decreased in leaves and stems, from 308 to 261 g kg<sup>-1</sup> DM and from 160 to 137 g kg<sup>-1</sup> DM, respectively. From first to third development stage, content of NDF in leaves and stems increased, from 380 to 514 g kg<sup>-1</sup> DM and from 537 to 590 g kg<sup>-1</sup> DM, respectively. The highest contents of ADF and hemicellulose were shown in plant parts at the third development stage. Three methods – acid detergent lignin (ADL), Permanganate lignin (PerL) and Klason lignin (KL) for determining lignin concentration in lucerne were compared. Each of these methods gave different lignin values for the same type of forage sample. For all samples, KL values were higher than ADL and PerL.

Keywords: lucerne, fibre, crude protein, lignin method

## Introduction

Lucerne (*Medicago sativa* L.) is an important forage crop because of its high nutritive value and high yield. Plant maturity is the major factor affecting morphology and determining forage nutritive value. The decline of forage nutritive value with age results primarily from a decrease in the leaf to stem ratio and the decline in nutritive value of the stem component. At early vegetative stage in lucerne, the crude protein (CP), neutral detergent fibre (NDF) and lignin contents in leaves were 297, 260 and 47 g kg<sup>-1</sup> DM, respectively. Stems at the same morphological stage contained 143, 753 and 72 g kg<sup>-1</sup> DM of CP, NDF and lignin, respectively (Paterson *et al.*, 1994).

The most commonly employed procedures for determining the lignin content fall into two categories: analytical methods that remove cell wall constituents except lignin and methods that oxidize the lignin polymer out of the cell wall matrix. These methods often give quite different values of lignin content (Fukushima *et al.*, 2000). The objective of this study was to investigate the major chemical constituents in leaf and stem of lucerne at three stages of maturity and to compare lignin content determined by different methods.

## Materials and methods

The experiment was designed as a randomized block with a 3 x 2 factorial arrangement of treatments replicated three times. In the first part of investigation three stages of growth of lucerne (cv K-28) were examined after first cutting. The first stage was cut on June 17 at full boot stage, another one on June 24 (around 40% flowering), and a third one in full flowering on July 1. The second factor was anatomical fraction; leaves and stems. Samples were hand cut with scissors at 5 cm height. Leaves and stems were separated and the samples were dried to constant weight at 65 °C for 48 h and dried samples were ground through a screen size of 1 mm. CP was measured by the Kjeldahl method modified by Bremner and Breitenbeck (1983). The NDF, acid detergent fibre (ADF) and acid detergent lignin (ADL) were isolated

as described by Van Soest (1963). Hemicellulose was calculated as the difference between NDF and ADF.

In the second part of the investigation, three methods of lignin analysis were compared at each of the three stages of growth identified above using the randomized block design. Permanganate lignin (PerL) determinations by Van Soest and Wine (1968) were performed on separate ADF preparations. Klason lignin (KL) was determined by the procedure of Theander and Westerlund (1986).

Data were analysed by analysis of variance. Effects were considered significant based on F-values ( $P < 0.05$ ). When a significant F-value was detected, pair-wise comparisons among means were conducted by using the LSD test.

## Results and discussion

Nutritive values of leaves and stems of lucerne at three stages of maturity are presented in Table 1. The greatest decrease of CP was noticed after the first growth stage in stems and after the second stage of growth in leaves. These data are in agreement with Babnik *et al.* (1994). The results show the importance of harvesting the plants at their early stages of growth and with a high nutritive value of leaves. Contents of cell walls increase differently in anatomical fractions of this legume species as the plant advances in age (Table 1). NDF and ADF concentrations increased to a greater extent in leaves than in stems, but the fibre concentrations were lower in leaves than in stems at all three stages of growth. A higher content of hemicellulose was found in leaves than in stems. There were significant differences between stage of growth and anatomical fraction ( $P < 0.01$ ) as well as interactions for CP, NDF, ADF and hemicellulose.

Table 1. Chemical composition in leaves and stems of lucerne ( $\text{g kg}^{-1}$  DM).

SG	Crude protein			NDF			ADF			Hemicellulose		
	Leaf	Stem	Av.	Leaf	Stem	Av.	Leaf	Stem	Av.	Leaf	Stem	Av.
I	308	160	234	380	537	459	133	424	278	247	113	180
II	304	138	221	452	569	510	150	436	303	302	113	208
III	261	137	199	514	590	552	205	456	321	309	154	232
Av.	291	145		449	565		163	439		286	127	
	SG	AF	SGxAF	SG	AF	SGxAF	SG	AF	SGxAF	SG	AF	SGxAF
<i>P</i>	***	***	***	***	***	***	***	***	***	***	***	0.002
LSD <sub>0.05</sub>	1.31	1.06	1.48	10.1	8.2	11.6	11.2	9.1	12.9	12.7	10.3	14.6

NDF – neutral detergent fibre, ADF – acid detergent fibre, SG – stage of growth, AF – anatomical fraction.

I stage – full boot, II stage – around 40% flowering, III stage – full flowering, Av. – average, \*\*\* –  $P < 0.001$ .

Table 2. Content of lignin in leaves and stems of lucerne determined by different methods ( $\text{g kg}^{-1}$  DM).

SG	Leaf				Stem			
	ADL	PerL	KL	Average	ADL	PerL	KL	Average
I	41.1	19.6	53.3	37.9	82.6	98.5	113	98.1
II	46.3	21.4	63.5	43.7	102	110	139	117
III	46.5	29.7	67.8	47.9	118	126	146	130
Average	44.6	23.5	61.5		101	111	133	
	SG	LM	SGxLM		SG	LM	SGxLM	
<i>P</i>	***	***	0.171		***	***	0.018	
LSD <sub>0.05</sub>	3.9	3.9			3.5	3.5	4.9	

ADL – acid detergent lignin, PerL – Permanganate lignin, KL – Klason lignin, SG – stage of growth, LM – lignin method.

I stage – full boot, II stage – around 40% flowering, III stage – full flowering, \*\*\* $P < 0.001$ .

Overall, as lucerne matured, lignin increased in both investigated anatomical fractions ( $P < 0.01$ ), with differences between stages of growth (Table 2). The highest content of lignin determined by different methods was found at the third stage of growth.

The various methods for determining lignin content resulted in different amounts of residues, with PerL values being consistently lower ( $P < 0.01$ ) than ADL and KL values for leaves (Table 2). The KL residue concentrations were on the order of 25% to 165% times greater than the ADL and PerL. All methods reflected maturity trends, although the magnitude of the change was different. It is generally considered that the major limitation of the KL method for forage samples is the inclusion of protein in the insoluble residue, resulting in artificially high lignin values (Fukushima *et al.*, 2000)

## Conclusion

The greatest decrease in CP of lucerne was noticed after the second stage of growth in leaves, and after the first stage of growth in stems. Increases in NDF and ADF concentrations with advanced maturities were greater in leaves than in stems. The KL method resulted in lignin concentrations that were greater than those produced by the ADL and PerL procedures. The leaf fraction had the lowest content of PerL.

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# The effect of botanical composition on mineral content in hay from extensively managed mountain grasslands

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## Abstract

The grasslands in the Pieniny National Park (West Carpathians) are managed in a traditional, extensive way to preserve typical plant communities. Management consists of late mowing without fertilization which may cause low fodder value. The paper presents results on the concentrations of Ca, Cd, Cu, K, Mg, Mn, Na, Pb, S, Zn in swards and the main factors that influence these contents. The highest variation was found in Cd content and the lowest in Zn content. For all microelements except Mn the highest correlation coefficients were calculated for the share contributed by herbs. The results demonstrated the high importance of dicotyledonous in improving the feed quality of extensively managed grasslands.

Keywords: mountains grasslands, microelements, extensive management

## Introduction

Traditional extensively managed mountain grasslands are very important for preserving biodiversity. However, decreasing cost-effectiveness of farming in the mountains leads to discontinuation of use of grassland. The situation is particularly disadvantageous in the protected areas where the aims are to protect semi-natural ecosystems that have been formed by farming, and are functioning due to farming practices. In the Pieniny National Park, grasslands cover over 18% of its area. Traditionally they were cut for hay, usually by the end of July, and received no fertilizers. This method of management leads to low forage value of the sward, particularly high fibre content (Zarzycki *et al.*, 2005). Late date of cutting may also contribute to a decline in forage value because some microelements are translocated to underground parts (McDowell, 1996). The aim of the work described in this paper was to assess the influence of botanical composition on the selected macro and microelement content in the fodder from extensively managed, multi-species mountain grasslands.

## Materials and methods

The investigations covered traditionally extensively managed grasslands in the Pieniny National Park (49°25'N, 20°24'E). They are situated in the forest zone at altitude of 610-950 m a.s.l. The substrata are usually weakly acid brown soils (pH 5.2-5.4) with low content of P (4.44-6.25 mg kg<sup>-1</sup> of soil) and high content of K (92-398 mg kg<sup>-1</sup> of soil). Mean annual air temperature in the Pieniny Mountains is from 6.3 to 3.0 °C and annual precipitation total between 690 and 850mm. Twenty-two research plots (of 100 m<sup>2</sup> each) were distributed randomly on the main complexes of clearings in a way that reflected the diversity of plant communities. Ten subplots (0.1 m<sup>2</sup>) were chosen from each plot. The sward was cut by the end of July 2003 after a majority of grasses had formed seeds. Botanical composition of samples was based on the botanical-and-weight analysis (separation of all species). Macro and microelement content in biomass was determined using ICP-AES technique.

## Results and discussion

Occurrence of 78 plant species, including 13 grass species, 9 legumes and 56 herb species, were found in the sward samples from the investigated grasslands. The number of species in samples varied from 12 to 31. The most common species were: slender crosswort *Cruciata glabra*, colonial bentgrass (*Agrostis capillaris*), cocksfoot (*Dactylis glomerata*), imperforate St. John's Wort (*Hypericum maculatum*), red fescue (*Festuca rubra*), germander speedwell (*Veronica chamaedrys*) and buttercups (*Ranunculus* spp.). Despite a similar species composition for a majority of samples, percentage by weight of species in samples was very variable. For instance, the share of colonial bentgrass present in all samples, ranged between 3% and 35%. There was a considerable proportion in many samples of: brownray knapweed (*Centaurea jacea*), red fescue, cocksfoot, zigzag clover (*Trifolium medium*) and masterwort (*Astrantia major*). Some species, like couch grass (*Agropyron repens*), mat grass (*Nardus stricta*), greater knapweed (*Centaurea scabiosa*) and mountain clover (*Trifolium montanum*) were present in large quantities but only in a few samples. The share of grasses ranged between 10.7% and 76.5%, legumes from 2% to over 30%, and herbs from 13.3% to 66.5%. Woodrushes and sedges were rather seldom, with the share ranging between 0 and 12.6%. The content of mineral components in samples revealed considerable diversity (Table 1), which is often observed in grassland swards (Trzaskoś 1998; Steinshamn *et al.*, 2004). The recommended content of micro and macroelements in forage depends on many factors, particularly on animal species but also on synergistic and antagonistic relations between the minerals. Cadmium concentrations proved to be the most diverse and in many samples exceeded the values considered to be toxic. The other analysed elements occurred in the sward in the quantities meeting farm animal nutritional requirements (McDowell, 1996; Falkowski *et al.*, 2001). Only copper concentrations were slightly too low, which is typical for the majority of grasslands in Poland.

Table 1. Statistical parameters of macro- and micro- element contents in swards from the Pieniny National Park (N = 22).

	Cd mg kg <sup>-1</sup>	Pb mg kg <sup>-1</sup>	Zn mg kg <sup>-1</sup>	Mn mg kg <sup>-1</sup>	Cu mg kg <sup>-1</sup>	Na mg kg <sup>-1</sup>	Ca g kg <sup>-1</sup>	K g kg <sup>-1</sup>	Mg g kg <sup>-1</sup>	S g kg <sup>-1</sup>
Mean	0.55	0.67	49.2	145.3	4.2	24.0	10.9	14.5	2.0	0.9
Minimum	0.13	0.32	27.4	68.8	1.1	12.1	4.1	9.5	1.2	0.4
Maximum	1.27	1.1	64.7	231.5	6.7	40.5	15.6	20.2	2.7	1.5
Stand. dev.	0.3	0.16	9.9	53.3	1.2	8.9	2.6	3.0	0.4	0.3
Coefficient of variability	55	24	20	37	28	37	24	21	21	32

The results have not revealed links between the share of species and element contents in the sward. It might have been due to the presence of a number of species with a small percentage, which affects only to a slight degree the chemical composition of the whole sample. It may be supported by a lack of correlation between the content of elements and the number of species, but occurrence of correlation of some elements content, and Shannon-Wiener diversity index, (Magurran, 1988), considering share of species in a sample (Table 2). However, the most important factor influencing chemical composition of samples was the percentage of grasses and herbs. A considerable share of herbs in a sample significantly increased the contents of a majority of mineral components in forage, which is a commonly noted phenomenon (Trzaskoś, 1998; Steinshamn *et al.*, 2004). Manganese was the only element whose concentration was not associated with botanical composition. The presence of a considerable share of herbs also increased crude protein content and decreased the fibre content in hay from the same area (Zarzycki *et al.*, 2005).

Table 2. Correlation coefficients between elements content in the sward and botanical composition of samples (N = 22). Coefficients marked with \* are significant ( $P = 0.05$ ).

	Cd	Pb	Zn	Mn	Cu	Na	Ca	K	Mg	S
Grasses	-0.39	-0.53*	-0.23	0.09	-0.59*	-0.32	-0.76*	-0.30	-0.53*	-0.52
Legumes	-0.15	0.16	-0.09	-0.26	0.20	-0.34	0.32	0.08	0.09	0.029
Herbs	0.51*	0.45	0.41	0.04	0.73*	0.53*	0.66*	0.42	0.53*	0.42*
Sedges	0.39	0.21	0.05	0.06	-0.14	0.28	0.24	-0.12	-0.18	-0.17
Number of species	-0.19	-0.33	-0.22	-0.02	-0.16	0.29	0.16	-0.35	0.15	0.16
Shannon's diversity index	0.06	0.48*	0.00	0.00	0.30	0.42*	0.60*	0.10	0.47*	0.45*

## Conclusions

The main factor influencing the mineral content in the hay was the share of herbs in the sward. The concentrations of selected elements in the herbage were sufficient to meet the nutritional requirements of extensively kept farm animals.

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# Characterization of water soluble proteins in some forage species and their effects on fermentation process measured by a gas production method

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## Abstract

Five species of forage, including alfalfa hay (*Medicago sativa*), white clover (*Trifolium repens*), sainfoin (*Onobrychis vicifolia*), fodder burnet (*Poterium sanguisorba*) and maize silage (*Zea mays indentata*) along with two grass species, *Lolium perenne* and *Glyceria plicata*, were used for determination of water soluble protein characteristics. Extractions of forage samples were performed with deionized water under laboratory conditions and the insoluble residues were oven-dried and, together with original samples, used for characterization of ruminal degradation parameters using gas production technique. The water soluble protein was precipitated using acetone or polyethylenglycol, and protein constitution was monitored by polyacrylamide gel electrophoresis. Results showed that the highest amount of soluble protein as a percentage of total protein was found for *Lolium perenne*, and the lowest percentage was found for clover ( $P < 0.05$ ). Soluble protein separation was not possible without using sodium dodecyl sulfate (SDS) in gel electrophoresis, and by using SDS different molecular weight proteins were found in the soluble fraction extracted from the forages. The results from the gas production technique showed no significant difference between the two groups of original forage samples or insoluble residue samples ( $P > 0.05$ ), but within each type of forage, water extraction can both reduce and increase gas production and the increased gas production may be ascribed to removal of anti-nutritional factors with the water-soluble fraction.

Keywords: forage, SDS-PAGE, soluble protein, gas production

## Introduction

Forages contain varying amounts of material extractable by water or buffer solutions. This material includes minerals, protein, organic acids, and various simple sugars and their short-chain polymers. Forage undergoes rapid and extensive degradation of protein after it has been harvested. The subsequent utilization of the forages by ruminants is affected by this protein degradation. It is evident that conversion of protein to NPN is a result of plant and microbial enzyme activity. The 'soluble' pool thus may play an important role in the early stages of forage digestion. The gas production technique is used more and more to predict (for preliminary *in-vitro* studies) the fermentative digestion of forage or rations in the rumen.

In the present study, characteristics of soluble protein were studied by extraction of the forage proteins and monitoring the composition of the extracted proteins by SDS-PAGE, and secondly, to evaluate the contribution of the soluble fraction to total gas production *in vitro*.

## Materials and methods

**Forage preparation.** Five species of forage including alfalfa hay (*Medicago sativa*), white clover (*Trifolium repens*), sainfoin (*Onobrychis vicifolia*), fodder burnet (*Poterium sanguisorba*) and maize silage (*Zea mays indentata*), along with two grass species, *Lolium perenne* and *Glyceria plicata* (GP), were used. The forage samples were oven dried at 60 °C for 48 h,



milled through a 0.9-mm screen and subjected to water extraction. Initial extraction trials were performed with deionized water under laboratory conditions. The soluble components were extracted from the forages by mixing forage samples with deionized water at 39 °C (100 ml total solvent per gram air-dried forage). The samples then were filtered in tared Gooch crucibles (porosity 40 to 60 mm) under light vacuum. The insoluble material was dried at 55°C for 48 h, weighed and stored for gas production test. The soluble fraction extracted from the forages was collected and subjected to protein precipitation using polyethylenglycol solution 20% (v/v) or pure acetone (-20°C). The precipitate protein was collected by centrifugation of the fluid (15,000 × g for 30 min.) and subjected to SDS-PEGE for monitoring protein profile.

**Gas measurements.** Triplicate samples of the unfractionated forages, and the insoluble fractions remaining from previous extraction, were incubated in test tubes with a nominal volume of 13 ml. The tubes contained 250 mg of air-dried sample, 1 ml of rumen fluid, 5 ml of McDougal saliva, and were mixed by vortex. The tubes were flushed with carbon dioxide for 3 min before they were sealed with rubber stoppers. Pressure readings in all tubes were recorded 2 hourly for 24 h. Mean gas production data from blanks were subtracted from the recorded gas production of the standard on all the substrates to get the net gas production values. For comparison of gas production (unfractionated forages vs. the insoluble fractions), a complete randomized design with three replicate was used (SAS, 2003).

## Results and discussion

Electrophoretograms of soluble protein in alfalfa, clover and sainfoin were more or less the same, but in fodder burnet two groups of protein were separated in SDS-PEGE, one group with higher than 97 kDa molecular weight and another group less than 21 kDa (Figure 1). In maize silage all soluble proteins showed higher than 21 kDa molecular weight, and in *Lolium perenne* no protein with molecular weight less than 97 kDa has been observed in electrophoretograms. Polyethylenglycol solution (20% v/v) could not precipitate protein in alfalfa, sainfoin, maize silage and *Lolium perenne*, but very low temperature pure acetone precipitated soluble protein in all forages in this experiment (Table 1).

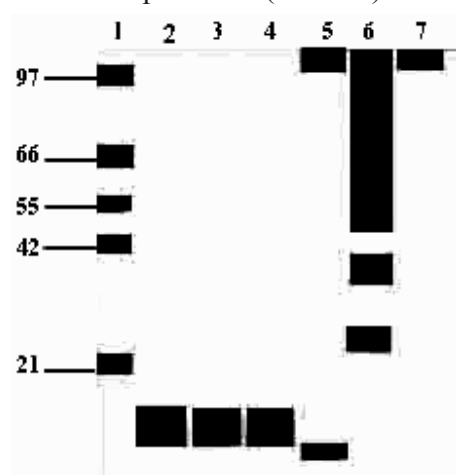


Figure 1. SDS-PEGE of acetone precipitated soluble protein in seven forage species. Lines one to seven are standards, alfalfa hay, clover, sainfoin, fodder burnet, maize silage, and *Lolium perenne*, respectively. The left axis shows standard molecular weight on a kDa basis.

The results in Table 1 show that the highest amount of soluble protein as a percentage of dry matter was found for fodder burnet and the lowest percentage was observed in maize silage, but as a percentage of total protein *Lolium perenne* has the highest solubility of protein and

the lowest percentage was found for clover. The data in Table 1 show the expected differences between legumes and grasses with respect to total protein content, and the expected trends in soluble protein content for non-legume forages.

The results from the gas production technique (Table 2) showed no significant difference between two groups of original samples or insoluble residue samples ( $P > 0.05$ ) but, within each type of forage, water extraction has both reduced and increased gas production, and the pattern is different for range species and sainfoin. The potential gas production is associated with degradability of the feed (Khazaal *et al.*, 1995). Therefore, the higher values obtained for the potential gas production in clover, sainfoin and maize silage might indicate a better nutrient availability for rumen microorganisms.

Table 1. The mean percentage of protein and water soluble protein in experimental forages.

	Forages				Maizesilage	<i>Lolium perenne</i>	GPF
	Alfalfa	Clover	Sainfoin	Burnet			
Total protein * (%)	16.30	19.00	16.70	16.72	7.90	8.60	8.70
Soluble protein* (%)	4.50	4.98	4.08	5.34	3.98	5.17	3.64
Polyethylenglycol □	-	+	-	+	-	-	+
Acetone □	+	+	+	+	+	+	+

GPF: *Glyceria plicata* fries; \*Total and soluble of protein as a percentage of dry matter.

□Negative sign shows no precipitation occurrence and positive sign shows vice versa.

It seems reasonable to assume that extraction of water-soluble protein would cause significant changes in gas production pattern, and also that forage digestion is affected by the presence of soluble components and their associated microbial population. In range forages, the soluble fraction (such as tannin) is a factor for inhibition of degradability, and when this fraction is absent from the forage, it is reasonable to expect a better degradation and a higher gas production (Table 2). In other forages (without anti-nutritional factor), water extraction will reduce availability of soluble protein and other soluble components as sugars and may influence microbial activity, and this aspect is the main reason for reduced gas production in extracted forages.

Table 2. The mean of gas production during 24 hours incubation of one-gram sample.

Kind of sample	Gas production (ml)							Average
	Alfalfa	Clover	Sainfoin	Burnet	Maize silage	<i>L. perenne</i>	GP	
Whole	237 <sup>a</sup>	301 <sup>a</sup>	262 <sup>b</sup>	230 <sup>a</sup>	267 <sup>a</sup>	265 <sup>b</sup>	222 <sup>b</sup>	255
Extracted	223 <sup>b</sup>	273 <sup>b</sup>	296 <sup>a</sup>	174 <sup>b</sup>	220 <sup>b</sup>	325 <sup>a</sup>	254 <sup>a</sup>	252
SE	3.31	4.09	3.35	3.40	4.07	3.31	3.35	3.57

GP: *Glyceria plicata*; Average indicate mean of gas production in whole forage or water extracted forage. SE: standard error. Means with the same letter in each column are not significantly different ( $P < 0.05$ ).

## Conclusion

Results from the gas production technique showed no significant difference between the two groups of original forage samples or insoluble residue samples, but within each type of forage, water extraction can both reduce and increase gas production. Increased gas production may be ascribed to removal of anti-nutritional factors with the water-soluble fraction.

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# The importance of timothy variety for feed quality development in regrowths of swards mixed with meadow fescue

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## Abstract

Small plot experiments with three varieties of timothy in mixture§ with meadow fescue were conducted during 2006 and 2007 at different sites in Norway. A first cut was taken simultaneously for all varieties at heading of 'Grindstad', and the yield and quality of the following regrowths were investigated when heat sums of 500, 700 and 850 day degrees (base temperature 0 °C) had been accumulated. On all sampling occasions of the regrowths, swards with 'Grindstad' returned higher yields with lower quality, higher proportion of generative shoots and lower proportion of meadow fescue than the swards with 'Noreng' and 'Vega'. Overall there was a high correlation between the proportion of generative shoots, DM yield and feed quality. The higher quality of swards with 'Noreng' and 'Vega' might both be related to their higher proportion of meadow fescue and their lower proportion of generative timothy shoots. The results imply that varieties that develop generative shoots in regrowths may be chosen if yield quantity is to be maximized. The rather steep decline in quality related to generative regrowths must also be taken into consideration when variety is chosen.

Keywords: digestibility, DM yield, NDF, INDF, *Phleum pratense*, summer growth

## Introduction

Most meadows in Norway are sown with a mixture of Norwegian varieties of timothy (*Phleum pratense* L.), meadow fescue (*Festuca pratensis* Huds.) and red clover (*Trifolium pratense* L.). The most commonly used variety of timothy, 'Grindstad', originates in Southern Norway while 'Noreng' and 'Vega' are bred in Northern Norway for high winter persistency. The origin of the varieties and the characteristics that are selected for during breeding will determine their growth pattern, yield potential and reactions to the environment, for instance to daylength. The higher DM yields obtained for 'Grindstad' than for the other two in variety trials (Molteberg and Enger, 2006) might be related to its higher rate of generative development in spring growth and also a high proportion of elongated shoots in regrowths. The difference in phenological development between the varieties may also differentiate them regarding competitiveness in mixed swards and in feed quality. The objective of the present work was to investigate the feed quality and yield development in regrowths of mixed swards of timothy and meadow fescue related to timothy variety.

## Materials and methods

The experiment was established at six sites in 2005 and three sites in 2006. Three trials were located in upland areas in Eastern Norway, four fields were situated in Central Norway and two trials were located in Northern Norway. Three varieties of timothy ('Grindstad', a southern type, 'Noreng' and 'Vega', both northern types) were seeded on small plots, in a mixture with 30% (weight basis) 'Norild' meadow fescue. The experimental fields were fertilized with compound fertilizer according to common practice in the region. In the first year of the ley a first cut was taken simultaneously for all varieties at heading of 'Grindstad', and the yield and quality of the following regrowths was investigated when heat sums of 450,

600 and 750 day-degrees (base temperature 0 °C) had been accumulated. On average, for nine trials, the actual heat sums were 502, 673 and 836 day-degrees, respectively. The developmental stage of timothy at harvest was assessed as 'Mean stage by count' (Moore *et al.*, 1991). The proportion of sown grasses was assessed visually as part of DM yield at each harvest. Dried samples were analysed using Near Infrared Reflectance Spectroscopy (NIRS) (Fystro and Lunnan, 2006). A measure of energy concentration (feed units (FU) kg DM<sup>-1</sup>) was determined according to Lunnan and Marum (1994). Data (means from each of nine sites) were subjected to ANOVA with variety (fixed) and site (random) as factors. Means for varieties were later separated according to a Ryan-Einot-Gabriel-Welsch multiple range test.

## Results and discussion

There were no significant differences in yield, or in quality, according to variety at the first cut. The DM yield of the first cut was on average 6,400 kg ha<sup>-1</sup> with an energy concentration of 0.86 feed units kg DM<sup>-1</sup>. The mean stage by count was 2.8 (no consistent differences between varieties were revealed), which more or less corresponds to early heading. The proportion of meadow fescue was 0.26 in plots with 'Vega' and 'Grindstad', and 0.29 where 'Noreng' was sown. At the second cut 'Grindstad' produced the highest DM yield on all sampling occasions, and the proportion of meadow fescue was about 0.10 units lower on plots with 'Grindstad' compared to plots with 'Noreng' and 'Vega' (Table 1).

Table 1. Dry matter yield of the second cut (kg ha<sup>-1</sup>) and proportion of meadow fescue for three varieties of timothy at three different harvest times. Averages of nine trials are given. Means marked with different letters were significantly different.

	Early 2 <sup>nd</sup> cut, 502 dd.		Medium 2 <sup>nd</sup> cut, 673 dd.		Late 2 <sup>nd</sup> cut, 836 dd.	
	DM yield	M. fescue	DM yield	M. fescue	DM yield	M. fescue
'Noreng'	1,940a	0.29a	3,160a	0.35	3,990a	0.33a
'Vega'	2,130a	0.29a	3,270a	0.31	3,950a	0.31a
'Grindstad'	2,530b	0.24b	3,980b	0.22	5,330b	0.22b
<i>P-value</i>	< 0.01	< 0.05	< 0.01	ns	< 0.01	< 0.01

The energy concentration was lower and the concentration of NDF and INDF higher in swards with 'Grindstad' than in swards with the other two varieties (Tables 2 and 3). The lower quality of 'Grindstad' than of 'Noreng' and 'Vega' swards may in part be explained by a higher proportion of generative shoots, as expressed by a higher mean stage by count (Table 2). Overall there was a high correlation between the proportion of generative shoots, DM yield and feed quality. The observed differences in feed quality of the mixed herbage according to timothy variety might also have been caused by different contents of meadow fescue (Table 1). This species may have a higher energy concentration than timothy in regrowths (Bakken *et al.*, 2008). In herbage from trials with a low proportion of meadow fescue in the regrowths, there was also significantly lower energy concentration in 'Grindstad' swards than in swards with the other varieties. The consequences of these results might be that regrowths in swards dominated by 'Grindstad' should be harvested early if feed quality is important. The effect of timing of the second harvest on winter survival of the sward was not investigated in this experiment. This must also be taken into consideration when decisions regarding timing of cuts and varieties are taken.

Table 2. Energy concentration (FU kg<sup>-1</sup> DM) and developmental stage MSC (Mean stage by count) at the second cut for three varieties of timothy at three different harvest times. Averages of nine trials are given. Means marked with different letters were significantly different.

	Early 2 <sup>nd</sup> cut, 502 dd.		Medium 2 <sup>nd</sup> cut, 673 dd.		Late 2 <sup>nd</sup> cut, 836 dd.	
	FU kg <sup>-1</sup> DM	MSC	FU kg <sup>-1</sup> DM	MSC	FU kg <sup>-1</sup> DM	MSC
'Noreng'	0.98a	1.41	0.93a	1.55	0.89a	1.77
'Vega'	0.96b	1.61	0.92a	1.58	0.90a	1.77
'Grindstad'	0.94b	1.73	0.87b	2.19	0.84b	2.30
<i>P-value</i>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01

Table 3. Content of neutral detergent fibre (NDF) and indigestible neutral detergent fibre (INDF) in g kg<sup>-1</sup> at the second cut for three varieties of timothy at three different harvest times. Averages of nine trials are given. Means marked with different letters were significantly different.

	Early 2 <sup>nd</sup> cut, 502 dd.		Medium 2 <sup>nd</sup> cut, 673 dd.		Late 2 <sup>nd</sup> cut, 836 dd.	
	NDF	INDF	NDF	INDF	NDF	INDF
'Noreng'	459a	36a	500a	48a	525a	61a
'Vega'	480b	45b	507a	53a	518a	63a
'Grindstad'	500c	49b	551b	73b	557b	95b
<i>P-value</i>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.05	< 0.01

## Conclusions

The results from nine field trials imply that varieties of timothy that develop generative shoots in regrowths may be chosen if yield quantity is to be maximized. The rather steep decline in feed quality related to generative regrowths must also be taken into consideration when variety is chosen.

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## Red clover for silage: management impacts on chemical composition in the season after sowing

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### Abstract

Management impacts on the chemical composition of red clover (*Trifolium pratense* L.) crops for silage were examined in Ireland. The crops were two diploid cultivars of red clover (Merviot and Ruttinova), each as monocultures or with perennial ryegrass (*Lolium perenne* L. cv. Greengold). In the first season after sowing, crops received 0 or 50 kg inorganic N fertilizer ha<sup>-1</sup> in March and the first harvest was on 2 June or 19 June. Subsequent harvests were taken 50, 94 and 191 days after 2 June or 44, 86 and 174 days after 19 June. Herbage crude protein (CP) content and preservation indices for ensilage [dry matter (DM) and water-soluble carbohydrate (WSC) contents, buffering capacity (BC)] were measured at each harvest. Red clover mixed with perennial ryegrass resulted in a higher WSC content but a lower BC and CP content than when in monoculture. The early first harvest resulted in a higher WSC content and lower CP, DM and BC for the first-cut herbage compared to the later first harvest. Crops with Merviot tended to have a higher WSC but a lower CP than those with Ruttinova. Spring application of N fertilizer had little impact on herbage chemical composition.

Keywords: red clover, perennial ryegrass, management, chemical composition

### Introduction

Red clover (*Trifolium pratense* L.) is widely regarded as a specialist forage crop that can be ensiled. The yield, quality and preservation of red clover crops for silage are likely to depend on several management factors (Frame *et al.*, 1998). In Ireland, the effects of cultivar, companion perennial ryegrass (*Lolium perenne* L.), N fertilizer and harvest schedule on the yield and digestibility of red clover crops during the first season after sowing were reported by O'Kiely *et al.* (2006). It was shown that the inclusion of ryegrass with red clover improved forage yield and digestibility. An early first harvest date improved digestibility at the expense of yield, and red clover cultivar affected annual yield. Spring application of inorganic N fertilizer resulted in no benefit. This paper reports the treatment effects on crop crude protein (CP) and three indices for pre-disposition to successful preservation as silage: herbage dry matter (DM) and water-soluble carbohydrate (WSC) contents and buffering capacity (BC).

### Materials and methods

A field experiment was conducted at the Teagasc Grange Beef Research Centre, Dunsany, County Meath, Ireland (53°30'N, 6°40'W, 92 m above sea level). A 2<sup>4</sup> factorial of treatments relating to the management of red clover crops was laid out in a randomized complete block (n = 4) design with 64 plots (each 10 m x 2 m). The crops were two cultivars of early flowering, diploid red clover (Merviot and Ruttinova; derived from different genetic sources), each sown (Sept. 2001) in monoculture or with perennial ryegrass (cv. Greengold). The crops received 0 or 50 kg inorganic N fertilizer ha<sup>-1</sup> in mid-March and had a first-cut harvest date of 2 June or 19 June. Subsequent harvests were taken 50, 94 and 191 days after 2 June or 44, 86 and 174 days after 19 June. All plots were harvested to a stubble height of 5 cm and received

22 kg P and 95 kg K ha<sup>-1</sup> after the first, second and third harvests, and double those rates after the fourth harvest. Harvested herbage samples were immediately frozen (-20 °C) until further processing. For each crop, herbage DM content was determined by oven drying a sub-sample at 98 °C for 16 h. A second sub-sample was dried at 40 °C for 48 h, milled (1 mm screen) and assayed for CP (LECO FP-428 N analyser; AOAC, 1990), WSC (anthrone method; Thomas, 1977) and BC (Playne and McDonald, 1966). Data were analysed using a General Linear Model that accounted for each of the four factors and all interactions.

## Results and discussion

Table 1. Main effects of management factors on herbage composition of red clover crops for each harvest (H) in the first year after sowing.

	H 1				H 2				H 3				H 4			
	DM	CP	WSC	BC	DM	CP	WSC	BC	DM	CP	WSC	BC	DM	CP	WSC	BC
Cultivar																
Merviot	166	159	73	456	147	197	65	516	136	223	51	522	249	248	72	425
Ruttinova	161	172	66	475	146	207	53	529	136	232	45	524	247	250	71	434
<i>P</i> value	0.182	0.003	0.211	0.069	0.702	0.029	0.008	0.185	0.921	0.185	0.054	0.876	0.725	0.613	0.808	0.192
Ryegrass																
Mono. <sup>1</sup>	163	184	50	490	146	216	51	551	134	246	46	562	239	274	65	471
Mixture <sup>2</sup>	163	147	89	441	147	188	67	495	139	210	50	485	257	224	77	389
<i>P</i> value	0.987	***	***	***	0.758	***	***	***	0.055	***	0.179	***	0.006	***	***	***
N fert.																
No	163	166	70	471	144	204	56	532	134	230	47	528	249	251	71	436
Yes	164	165	69	460	148	200	62	514	139	225	49	518	247	247	71	424
<i>P</i> value	0.906	0.897	0.773	0.245	0.232	0.320	0.183	0.071	0.124	0.473	0.681	0.495	0.817	0.179	0.950	0.097
First cut																
Early	150	159	89	443	168	190	75	526	138	216	55	482	249	250	63	435
Late	176	172	50	488	125	214	43	519	135	240	40	564	247	248	80	424
<i>P</i> value	***	0.003	***	***	***	***	***	0.485	0.350	***	***	***	0.703	0.625	***	0.127

DM = Dry matter (g kg<sup>-1</sup>); CP = Crude protein (g kg DM<sup>-1</sup>); WSC = Water-soluble carbohydrate (g kg DM<sup>-1</sup>); BC = Buffering capacity (mEq kg DM<sup>-1</sup>); \*\*\* *P* < 0.001; <sup>1</sup>Monoculture; <sup>2</sup> Red clover/ryegrass mixture.

There were significant (*P* < 0.05) main effects (Table 1) and some two-way interactions between management factors in their effects on herbage chemical composition. There were no significant three- or four-way interactions.

The inclusion of ryegrass with red clover improved the ensilability indices by generally increasing WSC and lowering BC at a relatively similar DM content (Table 1). Because red clover is a legume it had a higher concentration of CP and proportionately less WSC than ryegrass. Non-structural carbohydrates are used in the fermentation process during ensilage. Therefore, the addition of ryegrass as a source of WSC could be recommended for the preservation of red clover-based crops as silage. The addition of ryegrass also improved yield and digestibility (O'Kiely *et al.*, 2006) and the benefits of its inclusion are consistent with previous work (Frame *et al.*, 1998).

The impact of ryegrass on chemical composition depended on the harvest schedule. In particular, for the first cut, the increase in WSC due to ryegrass was larger and the decreases in CP and BC were smaller for the 2 June than 19 June harvest dates. These differences were possibly due to an increased proportion of lignified stem in the crops, but more so for ryegrass than red clover. This result and previous results for yield and digestibility (O'Kiely *et al.*, 2006) highlight the quantity versus quality dilemma in producing any temperate forage crop



for silage. Nevertheless, the results imply that the ensilability of the first cut crop would be improved by the earlier harvest date, particularly if ryegrass is included.

The effects of harvest schedule on CP and ensilability indices of subsequent cuts were artefacts of the different durations of regrowth and timing of cuts that were imposed during the rest of the growth season. While it is difficult to establish the effects and causes of the two harvest schedules after the first cut, there were differences in herbage chemical composition across the four harvests (Table 1). Based on these results, it is possible to rank the crops in terms of risk of not achieving lactic acid dominant silage fermentation in the order of Harvests  $4 < 1 < 2 < 3$ . In contrast, herbage CP contents were in the order Harvests  $1 < 2 < 3 < 4$ . Thus, although CP can be a major contributor to BC, other factors clearly contributed in this study.

In comparison to the effects of ryegrass and harvest schedule, red clover cultivar had a relatively minor impact on CP and ensilability indices. Merviot appeared easier to preserve as silage than Ruttinova because of a higher mean WSC content and lower mean BC (and CP) content at a similar DM (Table 1). The effect of cultivar was also expressed when ryegrass was included and was most obvious in the first two harvests of the growth season. For most indices, the differences between cultivars did not depend on timing of the first cut, although the increase in BC due to the later first cut was smaller for Merviot than Ruttinova crops. Merviot was likely to be easier to preserve although its annual yield could be lower (O'Kiely *et al.*, 2006).

There was no effect of the spring application of inorganic N fertilizer on mean CP content or ensilability indices at any harvest (Table 1). This result was consistent with the absence of yield benefits reported previously (O'Kiely *et al.*, 2006) and implies that neither red clover nor the ryegrass grown with red clover responded measurably to the applied N. This suggests that all crops were able to meet their N demands using alternative sources of N (e.g. N<sub>2</sub> fixation, soil N mineralization).

This experiment has demonstrated significant impacts of management factors on the yield, quality and ensilability of red clover crops. However, in general, the indices of ensilability (DM, WSC and BC) suggested that it would have been difficult to achieve good preservation with any of the crops unless extensive wilting and/or application of sufficient effective additive were employed in addition to the management practices identified in this study.

## Conclusions

Red clover mixed with perennial ryegrass generally resulted in a higher WSC content but a lower BC and CP content than when in monoculture. The early first harvest date resulted in a higher WSC content and a lower BC and CP content compared to the later first harvest. Crops with Merviot tended to have a higher WSC but a lower CP content than those with Ruttinova. Spring application of inorganic N had little impact on herbage chemical composition.

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# Effect of different tiller types on the accumulation and digestibility of the herbage mass of timothy (*Phleum pratense* L.)

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## Abstract

Three different tiller types (vegetative, vegetative elongating, generative) can be found in primary or regrowth timothy (*Phleum pratense* L.), but only little is known about their digestibility or contribution to the herbage yield. The occurrence of different tiller types and their corresponding nutritive values were studied during the progressive development of primary and regrowth swards in Maaninka, Finland during 2006-2007 using timothy (cv. Tammisto II) in three replicates. Primary growth was cut at three different developmental stages at 7-day intervals and regrowth 4, 6 and 8 weeks after optimal primary harvest. At each cut, a subsample of the sward was visually separated into three tiller types by the occurrence of nodes and inflorescence and, furthermore, into leaf, stem, inflorescence and senesced material fractions. In addition to vegetative and generative tiller types, the vegetative elongating tiller is an abundant type existing in timothy vegetation, especially in the regrowth. The leaf content explains well the changes in the D-value of the primary growth, but clearly less in the regrowth. The vegetative elongating tillers appear to differ from generative tillers in terms of herbage mass productivity, proportion of leaf material and D-value.

Keywords: tiller type, primary growth, regrowth, D-value, proportion of leaves

## Introduction

Timothy (*Phleum pratense* L.) is the most important perennial forage grass grown for silage and hay production in Scandinavia and is widely grown also in other cool and humid regions in Europe, Asia and North America (Bélanger *et al.*, 2006). Two characteristics of forage production have to be emphasized: pursuit for high herbage yield and concurrent desire for adequate nutritive value of herbage mass (HM). Quantitative changes in the primary growth (PG) herbage yield and digestibility are temporally reverse; according to Kuoppala *et al.* (2007), in PG timothy-meadow fescue swards the content of digestible organic matter (D-value) decreases at a rate of 5 g kg<sup>-1</sup> DM d<sup>-1</sup>. Despite higher leaf proportion (LWR) of regrowth (RG), the nutritive value of RG is typically poorer than that of PG, and predicting the milk production potential of RG silages has not been very simple (Kuoppala *et al.*, 2007). Generally, PG development begins with vegetative tillers (VEG) and later transition from VEG to generative tillers (GEN) is thought to be accompanied by internode elongation, subsequent true stem formation and decrease in LWR. However, timothy also has a third type of tiller, elongating vegetative tiller (ELONG) (Höglind *et al.*, 2005). At present, the proportion of these tiller types in timothy PG and RG and their nutritive values remain to be elucidated.

## Materials and methods

The experiment was conducted at the MTT Agrifood Research Finland, Maaninka Research Station (68°10'N, 27°18'E) during growing seasons 2006 and 2007. The experimental field was established in 2005 in three replicates with 14.6 kg ha<sup>-1</sup> timothy (cv. Tammisto II). In

2006 and 2007, the experimental field was fertilized for PG with 90, 13.5 and 22.5 kg ha<sup>-1</sup> and for RG with 90, 0 and 31.5 kg ha<sup>-1</sup> of N, P and K, respectively. DM yields (above stubble height 7 cm) were determined for PG cuts at 7-day intervals as the sward was booting (PG1), in early heading (PG2) and in full heading (PG3) and for RG cuts 4, 6 and 8 weeks after PG2 (RG1, RG2 and RG3, respectively). At each cutting, three subsamples of the sward (stubble height 0 cm) were primarily fractionated into three tiller type fractions (VEG, ELONG and GEN, even if totally or partially senesced), loose senesced and loose living material and other species. Tillers were classified as follows: 1) VEG: pseudostem, no visible/palpable nodes; 2) ELONG: true stem with nodes, no inflorescence; 3) GEN: true stem and visible/palpable inflorescence. Secondly, VEG, ELONG and GEN fractions were separately divided into living leaf blade, stem (including leaf sheaths and pseudostem, where present), inflorescence and attached senesced tissue. All fraction samples were dried at +60 °C for 40 h and determined for DM weights. Organic matter digestibility for all fractions was determined by the pepsin-cellulase method described by Nousiainen *et al.* (2003) and ash content by ashing the samples at +600 °C. Consequential D-values (digestible organic matter in DM, g kg<sup>-1</sup>) were then determined. Finally, HM and D-value for each tiller type were calculated based on the proportion and related D-values and DM concentrations of the fractions.

The data was first analysed by ANOVA combining PG and RG, but separating the years. The data were unbalanced due to biological reasons of all tiller types not being abundant at each cutting. Comparison of the D-values of ELONG and GEN was of primary interest. This was done by analysing these types separately by ANOVA and excluding PG1 and RG1. The harvest (PG or RG) x tiller type interaction was significant ( $P < 0.012$ ) and therefore the data were finally analysed for each harvest (PG and RG) and year. Finally, the relationship between D-value and sward characteristics was analysed by correlation analyses.

## Results and discussion

VEG HM production (DM kg ha<sup>-1</sup>) was marginal in PG in 2006 and 2007, but in RG it produced prominent amounts of HM, especially in RG1. The amount of ELONG HM was abundant in both years, and particularly in later cuts of RG it comprised most of the HM. GEN HM production was dominant in PG, but in RG it was low (Figure 1). In addition to tiller type HM, total PG HM included 473 and 522 and RG HM 639 and 397 kg ha<sup>-1</sup> DM loose senesced material in 2006 and 2007, respectively.

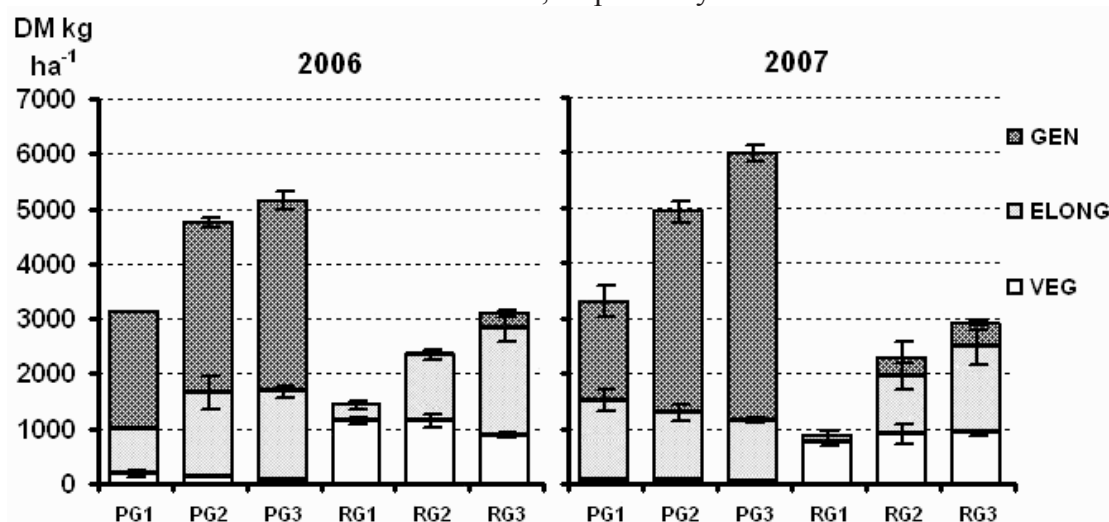


Figure 1. Dry matter (DM) yields (kg ha<sup>-1</sup>) of vegetative (VEG), elongating vegetative (ELONG) and generative (GEN) tiller types ( $n = 3$ ) during progressive development of primary growth (PG1-PG3) and regrowth (RG1-RG3) in 2006 and 2007. Loose senesced material not shown.

In VEG, the D-value was rather constant, at 678-697 g kg<sup>-1</sup> DM in PG and 673-714 in RG. This is logical since vegetative tillers have a high LWR. Furthermore, in VEG the stem fraction consists of pseudostems, not true stems. Therefore, comparison of D-value between ELONG and GEN is interesting, as they both have true stems. The D-value of ELONG and GEN was similar in PG, but in RG the D-value of ELONG was 38-40 g kg<sup>-1</sup> DM higher than that of GEN (Table 1).

Table 1. Effect of elongating vegetative (ELONG) and generative (GEN) tiller types on D-value (g kg<sup>-1</sup> DM) and leaf proportion (LWR) of herbage mass in primary growth (PG) and regrowth (RG). Values are means of two consecutive cuttings (PG2 and PG3 in PG and RG2 and RG3 in RG) ( $n = 6$  in each occasion except  $n = 5$  in PG GEN (<sup>a</sup>) in both years; effect of cutting time  $P > 0.05$  on each occasion).

		2006				2007			
		ELONG	GEN	SEM	<i>P</i>	ELONG	GEN	SEM	<i>P</i>
D-value	PG	640	636 <sup>a</sup>	1.4	0.15	671	665 <sup>a</sup>	7.6	0.33
	RG	723	683	3.3	0.01	669	631	5.4	0.015
LWR	PG	0.33	0.18	0.010	0.002	0.41	0.16	0.018	0.011
	RG	0.32	0.20	0.012	0.011	0.42	0.20	0.025	0.004

A higher D-value is expected if LWR is high. When analysed over the years, LWR correlated positively with the D-value, especially in PG. However, when each tiller type was analysed separately, in VEG there was no correlation between LWR and D-value ( $P > 0.05$ ) at neither harvest. In PG, a positive correlation existed in ELONG ( $r = 0.46$ ,  $P = 0.076$ ) and even a stronger one in GEN ( $r = 0.86$ ,  $P < 0.001$ ). However, in RG no correlation was observed.

The amount of attached senesced tissue of tillers may also explain the differences between tiller types. However, the proportion of attached senesced tissue was substantial only in VEG (mean  $\pm$  SD:  $0.17 \pm 0.10$ ), whereas in ELONG ( $0.06 \pm 0.04$ ) and GEN ( $0.04 \pm 0.02$ ) it was low. Therefore, the D-value decreased only in VEG with increasing proportion of attached senesced tissue and the relationship was not constant over the years. Therefore we assume that the digestibility of the true stem fraction in ELONG and GEN is different. This hypothesis will be tested as the research continues.

## Conclusions

In addition to the previously known vegetative and generative tiller types, the vegetative elongating tiller is an abundant type existing in timothy vegetation, especially in the regrowth. The leaf content explains well the changes in the D-value of the primary growth, but clearly less in the regrowth. The leaf proportion of the vegetative elongating tiller type is constantly higher than that of generative tillers. In the primary growth the D-values of the generative and the vegetative elongating tiller type appear to be quite similar, while in the regrowth the D-value of the vegetative elongating tillers is evidently higher.

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# Forage quality by animal fertilizer applications and by different grassland management

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## Abstract

A long-term small-plot trial was established in 2004 on grassland sites in the Rapotin locality. Treatments are applications of three fertilizers levels (no fertilization and cattle fertilizers) with three levels of intensity of cutting (2, 3 and 4 cuts per year). Three levels of fertilization correspond with stocking rate of 0.9, 1.4, and 2 LU ha<sup>-1</sup> (LU = livestock unit). Results obtained during two years of monitoring are reported. The chemical composition of the forage was evaluated on the basis of Czech state standard and *in-vitro* organic matter digestibility (OMD) is also reported. The concentration of crude protein (94-170 g kg<sup>-1</sup> DM) and the organic matter digestibility (61-72%) of the fodder were higher in treatments with higher intensity of cutting, with a significant decrease of concentration of crude fibre (310-230 g kg<sup>-1</sup> DM). The concentration of crude protein was also monitored.

Keywords: nutritive value, fertilization, cutting

## Introduction

In the Czech Republic an expansion of the permanent grassland areas is expected. In this situation it is necessary to identify the most suitable methods of grassland management for the future. Permanent grassland management for cattle breeding appears particularly attractive. It is important to provide the necessary quality of the feedstuffs to satisfy the animals' requirements. A major part of the Czech farms that are engaged both in crop and animal production utilize mineral fertilizers for this purpose. However, also grassland enables the effective utilization of natural fertilizers (manure, dung-water, slurry etc.).

## Materials and methods

A long-term small-plot trial was established in 2004 on permanent grassland sites in the locality of Rapotin, at 390-402 m a.s.l. Average values for temperature during the vegetation period (March-October) were 11.1 °C in 2005 and 12.2 °C in 2006, and the rainfall during the vegetation periods was 399.0 mm in 2005 and 460.2 mm in 2006. The grassland vegetation on the experimental stands was classified as *Arrhenatherion*. The dominant species in the permanent sward at the beginning of the trial were *Poa pratensis*, *Dactylis glomerata*, *Lolium perenne*, *Elytrigia repens*, *Taraxacum sect. Ruderalia* and *Trifolium repens*.

Experimental treatments simulating cattle grazing were as follows:

N0-2 – no fertilization, 2 cuts per year

N0-3 – no fertilization, 3 cuts per year

N0-4 – no fertilization, 4 cuts per year

Man 0.9-2 – cow manure + dung water with the load of 0.9 LU ha<sup>-1</sup> (which corresponds to 2 cuts per year and 54 kg N ha<sup>-1</sup>)

Man 1.4-3 – cow manure + dung water with the load of 1.4 LU ha<sup>-1</sup> (which corresponds to 3 cuts per year and 84 kg N ha<sup>-1</sup>)

Man 2.0-4 – cow manure + dung water with the load of 2.0 LU ha<sup>-1</sup> (which corresponds to 4 cuts per year and 120 kg N ha<sup>-1</sup>)

Slu 0.9-2 – slurry with the load of 0.9 LU ha<sup>-1</sup> (corresponds to 2 cuts year<sup>-1</sup> and 54 kg N ha<sup>-1</sup>)

Slu 1.4-3 – slurry with the load of 1.4 LU ha<sup>-1</sup> (corresponds to 3 cuts year<sup>-1</sup> and 84 kg N ha<sup>-1</sup>)

Slu 2.0-4 – slurry with the load of 2.0 LU ha<sup>-1</sup> (corresponds to 4 cuts year<sup>-1</sup> and 120 kg N ha<sup>-1</sup>)

The cow manure is applied in the autumn, dung water after the first cut; half of the slurry is applied in the spring and second half after the first cut. Nutrients in samples were analysed by the Czech State Standard 46 7092 (Testing methods for feeding-stuffs, 1985). Crude protein (CP) was determined by Kjeldahl procedure using a device Kjeltac Auto Distillation 2200 and ether extract (EE) by the Soxhlet method. The Fibertec System 2023 FiberCap (FOSS TECATOR) was used to analyse crude fibre (CF). Ash content was measured gravimetrically by igniting samples in a muffle furnace at 450 °C for 4 h. The *in-vitro* organic matter digestibility (OMD) was determined by the method of Tilley and Terry (1963) modified according Resch (1991). The ME (metabolizable energy), NEL (net energy of lactation), NEV (net energy of fattening), PDIE (ingested digestive protein allowed by energy), PDIN (ingested digestive protein allowed by nitrogen) was predicted by means of the regression equations for the organic matter digestibility (Pozdišek *et al.*, 2001) and by means of the equations mentioned in Petrikovič *et al.* (2000). The evaluation of the nutritive value in system NE, PDI is officially used in the Czech Republic and Slovakia. This system corresponds with the INRA system (Jarrige *et al.*, 1989). Statistical evaluation was made by analysis of variance (ANOVA).

## Results and discussion

The effect of different number of cuts on the evaluated parameters was statistically significant (Table 1).

Table 1 Chemical composition, digestibility and content of nutrients (mean of samples from 2005 and 2006).

AVG	FNtca [%]	CP [g kg <sup>-1</sup> DM]	CF [g kg <sup>-1</sup> DM]	OMD [g kg <sup>-1</sup> DM]	ME [MJ kg <sup>-1</sup> DM]	NEL [MJ kg <sup>-1</sup> DM]	PDIN [g kg <sup>-1</sup> DM]	PDIE [g kg <sup>-1</sup> DM]
N0-2	16.8	93.3	296.7	632	8.65	5.03	58.5	73.4
N0-3	21.5	116.7	263.8	680	9.25	5.44	73.2	80.5
N0-4	23.1	144.3	236.0	717	9.74	5.79	90.1	85.9
Man 0.9-2	21.1	125.1	266.5	634	8.63	4.98	77.9	80.6
Man 1.4-3	21.7	133.8	246.6	674	9.20	5.38	83.7	84.1
Man 2.0-4	22.7	169.0	223.8	682	9.21	5.38	104.7	88.1
Slu 0.9-2	22.4	128.3	267.7	642	8.77	5.08	80.0	81.7
Slu 1.4-3	22.1	138.0	263.7	653	8.93	5.19	85.9	83.3
Slu 2.0-4	22.5	169.9	232.1	677	9.12	5.33	105.2	87.5
ANOVA F – ratio								
Cutting	1.91	71.89**	29.49**	29.05**	15.08**	14.92**	72.95**	45.96**
Fertilization	1.72	14.16**	4.06*	2.53	1.95	2.53	14.13**	9.22**
Year	0.01	0.11	0.44	0.01	0.44	0.45	0.15	0.03

\*  $P < 0.05$  \*\*  $P < 0.01$ .

FNtca – nitrogen fraction soluble in trichloroacetic acid 10% w/v in water.

OMD – organic matter digestibility.

The tendency to decrease was found only in soluble nitrogen fraction (FNtca), particularly for variant N0-2 and N0-4. The results concerning contents of nutrients correspond with the results published previously (Pozdišek *et al.* (2006) and other authors).

For the optimal cattle nutrition it is suitable to use permanent grassland that has a PDIN/PDIE ratio of approximately 1. Treatments N0-4, Man 0.9-2, Man 1.4-3, Slu 0.9-2, and Slu 0.9-3

meet this requirement (Figure 1). Analogous to Pozdíšek *et al.* (2006) we can consider as suitable the treatments utilised with 3 cuts per year with a medium application of fertilizers.

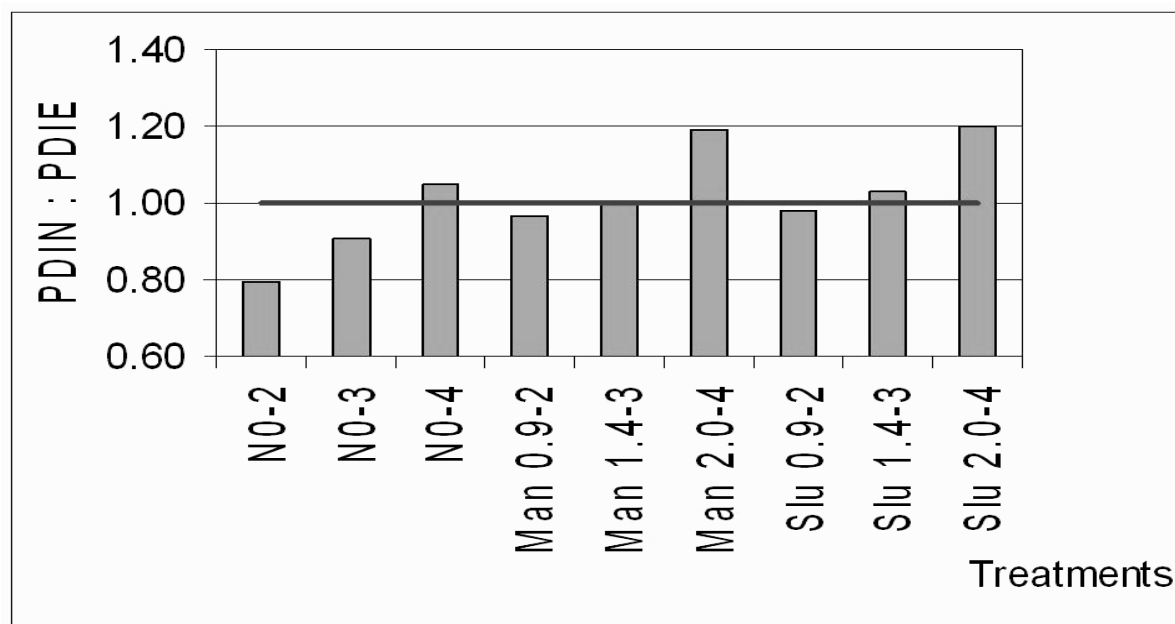


Figure 1. Ratio of ingested digestive protein allowed by nitrogen (PDIN): ingested digestive protein allowed by energy (PDIE), by treatments.

## Conclusions

It is possible to influence significantly the amount and the quality of the fodder by means of the grassland management, i.e. through the number of cuts and fertilization. Our findings presented in this paper are important for the nutrition of cattle and for efficient grassland management. Further research to enlarge the knowledge on this issue is still necessary.

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# Classification of mountain permanent grasslands based on their feed value

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## Abstract

Environmental and management conditions influence the floristic composition and yield of permanent grasslands. They can also influence their feed value. This study aimed to evaluate the variability of the feed value of the permanent meadows of the Massif Central of France and analysing the influence of the pedo-climatic factors and the management conditions on their feed value. A total of 182 plots were sampled at two dates along the first growth cycle during 2005. Grasslands were chosen according to the type of soil, (granite or volcanic) altitude (< 900 m; 900-1200 m; > 1200 m above sea level) and rainfall (< or > than 1000 mm per year). Fertilization and management of plots were estimated according to the information obtained from the farmers. The botanical composition was characterized for each sample. Chemical composition, organic matter digestibility and voluntary intake of samples were estimated by NIRS. By using multivariate techniques, plots were classified into six groups characterized by a gradient of digestibility. Variations in digestibility between groups can be explained from the contribution of specific grasses and forbs to the biomass. This study will contribute to develop a typology of permanent grasslands in the Massif Central.

Keywords: permanent grasslands, feed value, botanical composition, first growth cycle

## Introduction

Permanent grasslands play a fundamental role in the feed of herbivores. Different types of permanent grasslands can be distinguished based on the yield and on the botanical composition. However, a lack of knowledge of semi-natural grasslands' feed value has been reported (Bruinenberg *et al.*, 2002). Floristic composition of grasslands can influence their nutritive value because of differences in the chemical composition and digestibility of individual species, and also because the species of permanent grassland can vary in their phenology. The objective of this study was to characterize the variability of the feed value of permanent grasslands of the Central Massif and to relate it to their botanical composition, their management and their pedo-climatic conditions.

## Material and methods

One-hundred and eighty-two permanent grasslands located in the Massif Central of France were selected according to the type of soil (granite and volcanic), altitude (< 900 m; 900-1200 m; > 1,200 m above sea level), and rainfall (< or > than 1,000 mm per year). Each plot was sampled in 2005 at the date of making silage (around heading-stage of the more abundant grasses) and at the date of making hay (around flowering of the more abundant grasses) by farmers. Each date, five randomized sub-plots of 0.25 m<sup>2</sup> were cut within each plot at 5 cm above ground level. From the information provided by farmers, grassland fertilization was estimated and three levels (high; > 80 kg ha<sup>-1</sup>, medium; 40-80 kg ha<sup>-1</sup> and low; < 40 kg ha<sup>-1</sup>) were defined.

For each plot, a sub sample (125 g fresh material) was stored at -20 °C and was then separated into species by hand to assess the botanical composition. On dried and ground samples, crude ash (CA), crude protein (CP) neutral detergent and acid detergent fibre (NDF and ADF) contents, *in-vivo* organic matter digestibility (OMD) and voluntary intake in sheep (VI) were estimated by

near infrared reflectance spectroscopy (NIRS). The NIRS calibrations used are based on the INRA database and cover the same type of grasslands. Before predictions, the Mahalanobis distance (H) between the spectra of each sample and the average spectra of the calibration was calculated. Samples with  $H > 3$  were discarded. The SEP values ( $\text{g kg}^{-1}$ ) range between 9.6 for CP and 14.3 for NDF and they are of  $2.4 \text{ g g}^{-1}$  and  $4.9 \text{ g kg}^{-1} \text{ BW}^{0.75}$  for OMD and VI respectively. The N, P, K nutritional status was calculated according to Th  lier-Huch   *et al.*, (1999).

For each sampling date, a classification of the permanent grasslands was performed by means of multivariate analyses (correspondence analyses and hierarchical classification) on the dataset, gathering the pedo-climatic, the botanical composition, the chemical composition and the feed value variables. An ANOVA analysis followed by the Tukey test was performed in order to test possible differences between groups.

## Results and discussion

A great variability was evident for all the variables. In particular, OMD varied between 0.52 and 0.84 at the heading stage and between 0.50 and 0.74 at the flowering stage. The dry matter (DM) yield showed also a great variability (between 2.7 and  $5.4 \text{ Mg DM ha}^{-1}$  at the 'silage' stage. It was closely related to the level of fertilization. The multivariate analyses allow us to classify the grasslands into six groups, which could be characterized by their floristic composition and by the pedo-climatic conditions too. As the classification is very similar for the two sampling dates, we present the results of the 'silage' sampling date.

Groups G1 and G2 were located in areas where annual rainfall exceeds 1,000 mm ( $R^+$ ). The first group is strongly fertilized ( $F^+$ ) and the second one, received a moderate fertilization ( $F^-$ ). Plots of G1 are characterized by a high DM yield ( $> 5 \text{ Mg DM ha}^{-1}$ ) and the highest OMD of all groups (0.77) (Table 1). This group is also characterized by the high proportion of forbs (0.33) in the biomass (Table 2). Plots of G2 are characterized by a significantly lower DM production and OMD coefficient which agrees with the lower level of fertilization received.

Table 1. Yield ( $\text{Mg DM ha}^{-1}$ ), *in-vivo* organic matter digestibility (OMD  $\text{g g}^{-1}$ ), voluntary intake (VI) ( $\text{g kg}^{-1} \text{ BW}^{0.75}$ ), crude protein (CP) ( $\text{g kg}^{-1}$ ), and cell wall constituents (NDF and ADF) ( $\text{g kg}^{-1}$ ) of the six groups of permanent grasslands.

	Groups of permanent grasslands					
	G1/ $F^+R^+A^-$	G2/ $F^-R^+A^-$	G3/ $F^+R^-A^-$	G4/ $F^-R^+A^+$	G5/ $F^-R^-A^-$	G6/ $F^-R^-A^+$
N	36	24	70	4	35	11
Yield	5.01 a	3.41 b	5.14 a	5.40 a	3.85 ab	2.74 b
OMD	0.77 a	0.71 b	0.72 b	0.69 bc	0.70 b	0.62 c
VI	72.2 a	68.2 ab	71.7 a	71.1 ab	67.2 b	60.6 c
CP	124 a	122 a	115 ab	138 a	100 b	129 a
NDF	512 c	551 bc	562 b	554 bc	568 b	628 a
ADF	272 c	291 b	293 b	300 b	303 b	339 a

N: Number of plots;  $F^+$ : High level of fertilization ( $> 80 \text{ kg ha}^{-1}$ );  $F^-$ : Medium level of fertilization ( $40\text{--}80 \text{ kg ha}^{-1}$ );

$F^-$ : low level of fertilization ( $< 40 \text{ kg ha}^{-1}$ );  $R^+$ : rainfall  $> 1000 \text{ mm}$ ;  $R^-$ : rainfall  $< 1000 \text{ mm}$ ;

$A^-$ : less than 900 m;  $A^-$ : 900 m to 1200 m;  $A^+$ : more than 1200 m above sea level.

Averages without common letters on the same line differ significantly ( $P < 0.05$ ).

Group G3 includes meadows with an annual rainfall of less than 1000 mm ( $R^-$ ) and high nitrogen fertilization ( $F^+$ ). The DM yield of plots in G3 is similar as in G1, but the OMD coefficient is significantly lower. Meadows of G3 have the highest proportion of grasses (0.84) (Table 2).

The meadows in group G4 are situated in a high rainfall area ( $R^+$ ) and receive a moderate fertilization ( $F^-$ ). Their OMD is intermediate and they present a high proportion of forbs (0.40) and the lowest proportion of grasses (0.55) (Table 2). They differ from G2 because they are located at very high altitude ( $> 1200 \text{ m}$ ). The last two groups (G5 and G6) are located in areas  $R^-$ , and they are moderately or very little fertilized. They are characterised by low values of

digestibility (0.70 and 0.62 respectively). G6 shows a very high proportion of senescent material (0.33).

Table 2. Contribution of grasses, forbs, legumes and senescent material to the total biomass. The contribution of the most representative species is indicated for grasses and forbs.

	Groups of permanent grasslands					
	G1/F <sup>+</sup> R <sup>+</sup> A <sup>=</sup>	G2/F <sup>-</sup> R <sup>+</sup> A <sup>=</sup>	G3/F <sup>+</sup> R <sup>-</sup> A <sup>-</sup>	G4/F <sup>-</sup> R <sup>+</sup> A <sup>+</sup>	G5/F <sup>-</sup> R <sup>-</sup> A <sup>±</sup>	G6/F <sup>-</sup> R <sup>-</sup> A <sup>±</sup>
Grasses	0.61 bc	0.64 bc	0.84 a	0.55 bc	0.73 b	0.59 bc
<i>Dactylis glomerata</i>	0.14 a	0.04 b	0.15 a	0.06 ab	0.07 ab	0.05 ab
<i>Festuca rubra</i>	0.01 b	0.18 a	0.03 b	0.15 ab	0.12 a	0.03 b
<i>Holcus lanatus</i>	0.06 b	0.02 b	0.06 b	0.00 b	0.15 a	0.04 b
<i>Lolium perenne</i>	0.07 ab	0.04 b	0.11a	0.00 b	0.06 ab	0.00 b
Forbs	0.33 a	0.25 ab	0.11 c	0.40 a	0.20 b	0.08 c
<i>Polygonum bistorta</i>	0.01 b	0.01 b	0.00 b	0.33 a	0.00 b	0.00 b
<i>Taraxacum spp.</i>	0.15 a	0.02 b	0.04 b	0.01 b	0.04 b	0.00 b
Legumes	0.04 ab	0.07 a	0.02 b	0.01 b	0.03 ab	0.00 b
Senescent material	0.02 b	0.04 b	0.03 b	0.04 b	0.03 b	0.33 a

The 6 groups differ also by the species composition within the botanical families (Table 2). *Lolium perenne*, and *Dactylis glomerata* are the most abundant species of G1 and G3. On the other hand, high proportions of *Festuca rubra* were found in G2, G4 and G5, and *Holcus lanatus* was the main grass species in G5. Concerning the forbs, *Taraxacum spp.* was the more abundant species in G1 whereas *Polygonum bistorta* accounted for 82% of the total forbs proportion of G4. The more representative species of G5 were *Centaurea nigra* and *Plantago lanceolata*. The presence of species such as *Fraxinus excelsior* characterizes G6.

Differences in botanical composition are consistent with those in nutritive value, intake and chemical composition. The presence of high quality species like *Taraxacum spp.* can explain the highest digestibility in G1 (Schubiger *et al.*, 2001). The presence of lower quality grasses like *Festuca rubra* (Pontes *et al.*, 2007) in G2, G4 and G5 and of lower quality forbs like *Polygonum bistorta* in G4 can explain the lower digestibility in these groups.

## Conclusions

The results of this study will help to define different types of permanent grasslands linking pedo-climatic conditions and management with the botanical composition, the chemical composition and the feed value.

## Acknowledgements

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# Digestibility and fibre fractions of perennial ryegrass (*Lolium perenne* L.) as affected by cultivar and ploidy level

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## Abstract

Two field trials were carried out at 3 sites in northern Germany during the 2006 growing season to investigate the effect of genotype (trial 1) and ploidy level and cutting regime (trial 2) on the digestibility and fibre fractions of perennial ryegrass. In trial 1, the 20 evaluated genotypes showed relevant variations, e.g. a difference of 63 g NDF kg<sup>-1</sup> DM was observed in the first cut between the highest and lowest values. In trial 2, significant differences between diploids (2n) and tetraploids (4n) were recorded in few of the studied ploidy families, e.g. the 2n genotypes gave higher values than their 4n isogenic lines in the first cut and annual average neutral detergent fibre and acid detergent fibre contents. For digestible organic matter and metabolizable energy, the trend between the two studied ploidy levels was unclear.

Keywords: perennial ryegrass, genotype, ploidy level, digestibility, fibre

## Introduction

In temperate environments, perennial ryegrass (*Lolium perenne* L.) is the most widely used species in grassland because of its high palatability, digestibility and persistence. For a more sophisticated evaluation of forage quality other parameters should be considered which allow a better assessment of the carbohydrate degradation processes in the rumen. The main aim of the current project is to screen systematically a set of perennial ryegrass genotypes in terms of their yielding potential, as well as a process-oriented assessment of their carbohydrate contents, in order to quantify the impact of genotype and ploidy level. In this paper, results from the first growing season regarding fibre fractions, digestible organic matter (DOM), and metabolizable energy (ME) content are presented and discussed.

## Materials and methods

Two field trials were conducted at three sites in northern Germany during the growing season of 2006. In the first trial, 20 diploid intermediate heading genotypes were screened in a randomized complete block design with three replicates in a 4-cut system. A lattice design was used in the second trial to evaluate 25 genotypes under two cutting regimes, A and B. These 25 genotypes (20 *Lolium perenne*, 3 *L. multiflorum*, and 2 *Festuca pratense*) belonged to 9 ploidy families, each consisting of one diploid (2n) genotype and the tetraploid (4n) near isogenic line(s) derived from it. The *L. multiflorum* and *F. pratense* genotypes were mainly used for the purpose of comparison. Cutting regime A was applied at ear emergence of the earliest genotype, with a cutting interval of 5 weeks. Cutting regime B began 2 weeks later than regime A with an interval of 6-7 weeks. Forage quality was estimated by NIRS, based on the following wet chemical analysis: Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF) using the semiautomatic ANKOM apparatus (Van Soest *et al.*, 1991). DOM (g kg<sup>-1</sup> DM) and ME (MJ kg<sup>-1</sup> DM) were calculated according to Weißbach *et al.* (1999) based on the cellulase method. Data were statistically analysed using the mixed procedure of SAS, with the least significant difference procedure for mean comparison, and probabilities being adjusted by Tukey test (main effects) and Bonferroni-Holm test (interactions).

## Results and discussion

**Trial 1:** NDF, ADF, ME values varied significantly among the 20 tested genotypes and also among the 3 sites in both the first cut and the annual average. The DOM showed significant variations among the 20 genotypes in the first cut, the annual average and among the 3 sites only in the first cut. A significant difference of about 63 and 44 g NDF kg<sup>-1</sup> DM was obtained between the highest and lowest values from the first cut and annual average, respectively. Similarly, a difference of 51 and 38 g ADF kg<sup>-1</sup> DM, 68 and 33 g DOM kg<sup>-1</sup> DM, 0.98 and 0.58 MJ ME kg<sup>-1</sup> were achieved from the first cut and the annual average, respectively.

**Trial 2:** Analysis of variance (Table 1) revealed significant main effects of sites, genotypes and cutting regimes on the studied quality parameters in the first cut, while the 3-way interaction did not show any significant effect. However, the 3-way interaction exerted a significant influence on all the four quality parameters in the annual average.

Table 1: Analysis of variance for first cut and annual averages (A.A.) for NDF, ADF, DOM and ME for the genotypes in the three sites under the two cutting regimes.

Effect	df	Pr > F							
		NDF		ADF		DOM		ME	
		1 <sup>st</sup> cut	A.A.	1 <sup>st</sup> cut	A.A.	1 <sup>st</sup> cut	A.A.	1 <sup>st</sup> cut	A.A.
Site (S)	2	0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Cut Regime (R)	1	0.9324	<0.0001	0.0321	0.0150	0.0018	<0.0001	<0.0001	<0.0001
S x R	2	0.0847	<0.0001	0.2446	0.0002	0.0132	<0.0001	0.0004	<0.0001
Genotype (G)	24	0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0002
S x G	48	0.0023*	0.0016	<0.0001	<0.0001	<0.0001	0.0053	0.2153	0.0106
R x G	24	0.1130	<0.0001	0.0508	<0.0001	0.1965	0.0002	0.7208	<0.0001
S x R x G	48	0.3265	0.0007	0.5035	0.021	0.5142	<0.0001	0.9972	0.0001

\*non significant after applying Bonferroni-Holm test.

Since the main aim of trial 2 is to highlight the impact of ploidy level, only the significant interactions, including genotypes, will be clarified and discussed. Figure 1 shows significant differences between 2n and 4n in the first cut. The 2n genotypes in families A, E and I gave significantly higher values than their 4n derivatives with regard to NDF and ADF. The situation was reversed in the case of DOM, where the 4n of the families A, E and F showed higher values than the 2n of the same families. The trend was not clear in the amount of ME produced, the 4n of family D were better than the 2n of the same family, while the 2n of family H produced more ME than the 4n. Concerning the annual average, the significant influence of the 3-way interaction was clear in few ploidy families. For NDF, 2n genotypes of families E, F and I were higher than their 4n derivatives. The same trend was observed in family I for ADF (Figure 2a). For DOM, a significant interaction was observed only in two ploidy families; in family A the highest values were in favour of the 4n genotype, but in family F the 2n genotype showed higher DOM (Figure 2b). In families E and F the 2n genotypes gave higher values than the 4n ones in the amount of ME produced, while the situation was reversed in the Family A (Figure 2c). Several studies have reported a slight superiority of the 4n genotypes when compared to the 2n genotypes with respect to the DOM and the different quality aspects (Boller, 1999; Gilliland *et al.*, 2002). In contrast, Nekrosas (2002) observed no significant differences between 4n and 2n genotypes. However, the unclear trend of 2n and 4n observed in this study may be attributed to the genetic makeup of the near isogenic lines incorporated in the study, as the effect of 4n on quality parameters is dependent on the genetic background of the genotypes (Smith *et al.*, 2001).

## Conclusions

Results obtained from this investigation indicated relevant differences in the tested quality parameters within the intermediate heading genotypes. With respect to ploidy level, the unclear



tendency does not allow a definite conclusion to be drawn. More attention should be paid to the genetic constitution of the genotypes under comparison. Work will be completed by investigating further quality parameters, and the same will be done for the second experimental year.

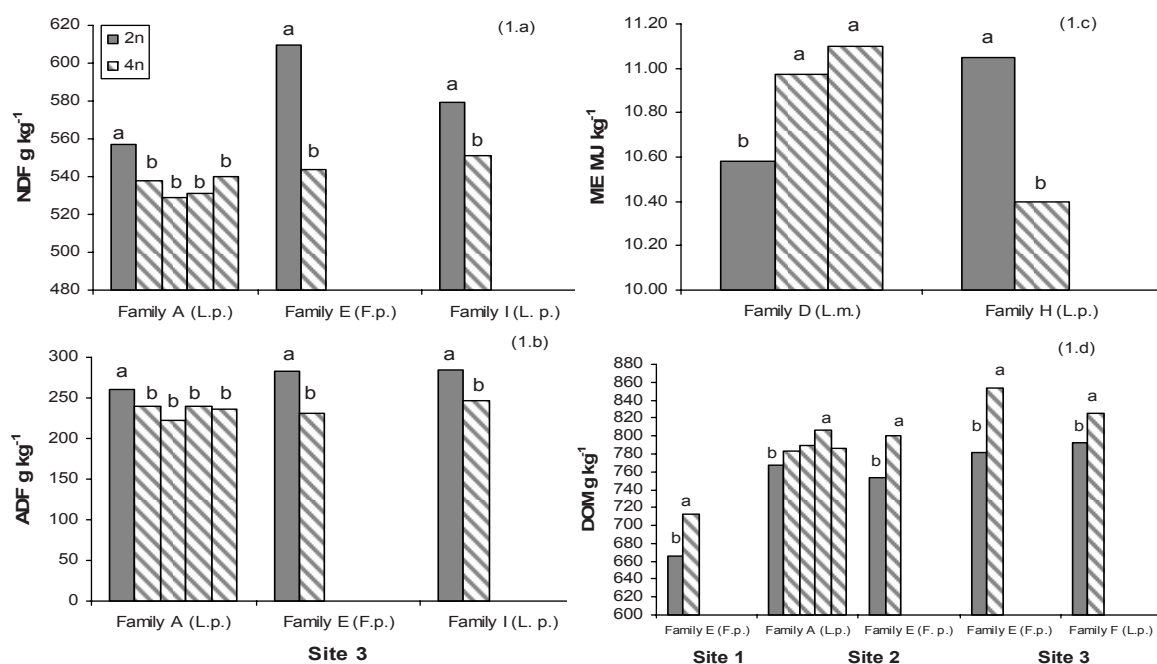


Figure 1. Differences in 1<sup>st</sup> cut NDF g kg<sup>-1</sup> DM (1.a), ME MJ kg<sup>-1</sup> (1.b), ADF g kg<sup>-1</sup> DM (1.c) and DOM g kg<sup>-1</sup> DM (1.d) between diploids and tetraploids within the same ploidy family.

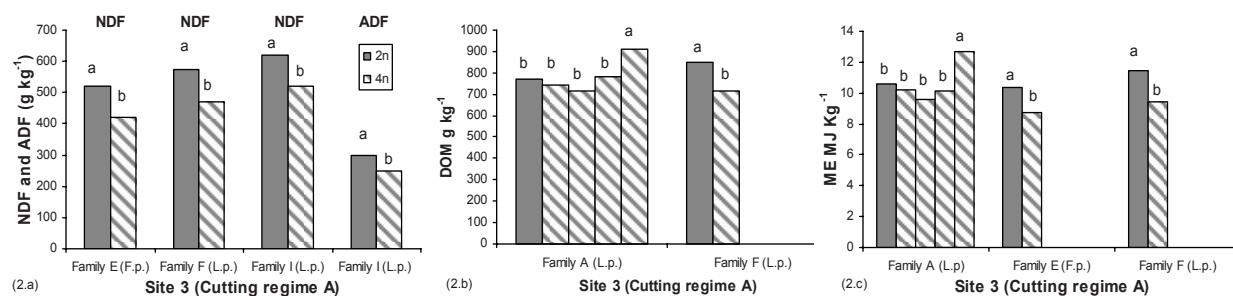


Figure 2. Differences in annual NDF, ADF g kg<sup>-1</sup> DM (2.a), DOM g kg<sup>-1</sup> DM (2.b) and ME MJ kg<sup>-1</sup> (2.c) between diploids and tetraploids within same ploidy family as affected by the 3-way interaction.

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## Changes in the non-structural carbohydrate content of two cool-season steppe grasses under different cutting frequencies

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### Abstract

Overgrazing, as a result of land use intensification, is a serious problem in the semi-arid grassland steppes of Inner Mongolia, China. The objective of the presented study is to characterize grassland degradation processes. Plant growth analysis and herbage quality parameters were used to describe short-term responses of a *Stipa grandis*/*Leymus chinensis*-community subjected to different defoliation intensities. Non-structural carbohydrates (NSC) play an important role in regrowth of defoliated grasses and are assumed to decrease when defoliation frequency increases.

A cutting-frequency experiment was conducted in the natural grassland of Inner Mongolia. Herbage samples of the two dominant species, obtained over the vegetation periods of 2004 and 2005, were analysed for glucose, fructose, sucrose, fructan and starch using Ion-Exchange-Chromatography on a sub-sample, with subsequent Near-Infrared Spectroscopy (NIRS) estimation. Mean NSC-contents decreased in frequently cut treatments in 2004, but not in 2005. These differences are probably a result of climatic variation. The composition of the NSC differed between species. After two experimental years it can be stated that defoliation influences the NSC-content, but the parameter is more susceptible to climatic variation than to defoliation.

Keywords: *Leymus chinensis*, *Stipa grandis*, cutting frequency, non-structural carbohydrates, semi-arid steppe

### Introduction

Land use intensification by enhancing the number of grazing animals is a serious problem in the semi-arid grassland steppes of Inner Mongolia, P.R. China. Since 2004 the Sino-German MAGIM-project (Matter fluxes in grasslands of Inner Mongolia as influenced by stocking rate, [www.magim.net](http://www.magim.net)) investigates this problem and the resulting agronomical and ecological consequences in a multidisciplinary approach. As a sub-project of MAGIM the working group Grassland and Forage Science/Organic Agriculture, University of Kiel and the Institute of Botany, Chinese Academy of Sciences, Beijing are working together in identifying short-term responses to defoliation on plant and community level to characterise the onset of grassland degradation processes. In a cutting-frequency experiment the grassland sward and specifically the dominant species *Stipa grandis* and *Leymus chinensis* were studied. *Stipa grandis* is a tussocky needle grass flowering in early to mid September and *L. chinensis* is a rhizomatous grass, belonging to the genus of wild-ryes flowering in mid June. Both grasses are perennials of the C3-type. Part of the concept was the analysis of non-structural carbohydrates (NSC) as they play an important role in the metabolism of grasses and are directly influenced by defoliation, as the NSC source, green aboveground biomass, is removed. The hypothesis is that grass plants respond directly to increasing defoliation frequency with decreasing NSC contents, also in semi-arid grasslands. The sugars glucose, fructose and sucrose, as well as the



reserve carbohydrates fructan and starch were analysed in aboveground biomass of the two dominant species over the vegetation periods of 2004 and 2005.

## Materials and methods

A completely randomized block design with 4 replications was used to test 3 different cutting frequencies: treatment I (T I) was cut once at the end of the growing season, T II was cut every 6 weeks and T III was cut every 3 weeks. The swards were cut to 2.5 cm stubble height. Biomass samplings to study the dry matter (DM) increment were conducted in T I and T II every 2 weeks in 2004 and every 6 weeks in 2005, whereas T III was consistently sampled every 3 weeks in both years. The sampling period started in the first week of June and the cutting intervals started in the first week of July. Sampling and cutting were finished around 10th September for all treatments in both years. At sampling dates the above-ground biomass was clipped by hand to 1 cm stubble height and was afterwards divided into four groups: *S. grandis* and *L. chinensis*, the necrotic material and other remaining species (4 quadrats per plot, each sized 0.25 m<sup>2</sup>). Samples were dried for 24 h at 60 °C, and ground to 1 mm. In Kiel all samples were scanned by a Near-Infrared-Spectrometer (NIRS) and a sample sub-set was analysed for NSC by high pressure anion exchange chromatography with pulsed amperometric detection (HPAEC-PAD). Sugars and fructans were extracted by cold water extraction. Afterwards fructan was hydrolysed by 2N HCl and incubated at 80 °C for 2 h. Starch was extracted from the dried pellet using amyloglucosidase (*Aspergillus niger*). Sugars were then measured by HPAEC-PAD and fructan and starch content calculated. Thereafter NSC contents were estimated by NIRS for all samples.

Annual means of NSC contents were subjected to analysis of variance, separately for *S. grandis* and *L. chinensis* and for the two experimental years. 'Replication' and 'treatment' were included into the statistical model as fixed factors. With significant F-value ( $P < 0.05$ ) means were tested by Student's t-test and probabilities corrected by the Bonferroni-Holm test.

## Results and discussion

For NSC-content the factor 'treatment' was significant for *S. grandis* in 2004 and 2005, whereas this factor was only significant in 2004 for *L. chinensis*. The mean cumulative DM yield of the community was 250 g DM m<sup>-2</sup> in 2004 and 120 g DM m<sup>-2</sup> in 2005. The decrease in 2005 was caused by low precipitation, as only 166 mm of rain were recorded in 2005, but average rainfall in a normal year is 350 mm at this site. *S. grandis* accounted on average for 33% and *L. chinensis* for 26% of the DM yield of the investigated community in 2004 and 2005. Figure 1 presents the NSC content separately for both species and both years, indicating the composition of NSC. Glucose and fructose are primary end-products of photosynthesis. The content of these monosaccharides, which have mainly metabolic functions in the cell, were reduced in *S. grandis* in both years, whereas the glucose and fructose content of *L. chinensis* was rather constant and only slightly influenced by defoliation frequency. The disaccharide sucrose serves as transport form for carbohydrates in the plant. It has been suggested that high sucrose concentrations in plant tissue of grasses induce the synthesis of fructan, but Chatterton *et al.* (1989) were not able to confirm this hypothesis for all grass species they tested. The two investigated grass species differ markedly from each other in sucrose content. While the sucrose content of *S. grandis* never fell below 25 mg g<sup>-1</sup> DM, the sucrose content of *L. chinensis* never exceeded 18 mg g<sup>-1</sup> DM. Sucrose content was significantly reduced in frequently defoliated treatments in *S. grandis*, while for *L. chinensis* differences due to defoliation were only observed in T III in 2005. These differences in sucrose content can be accounted for as species specific.

The main reserve carbohydrate in C3-grasses was often reported to be fructan (McIlroy, 1967), but also starch has reasonable relevance as reserve carbohydrate in temperate C3 grasses as observed by Chatterton *et al.* (1989).

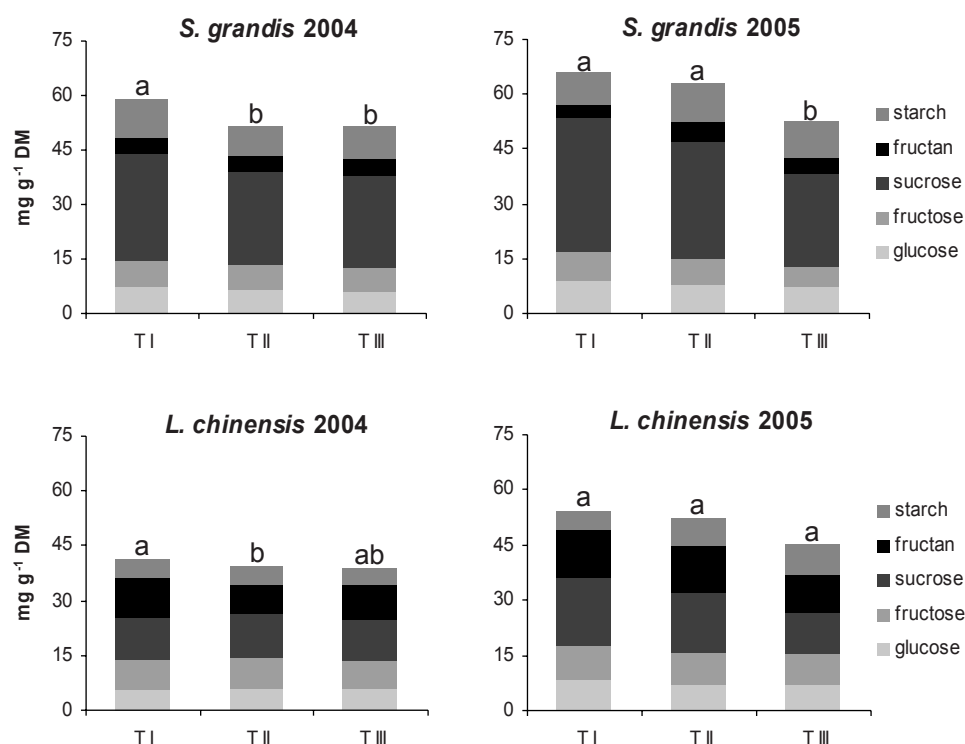


Figure 1: Mean non-structural carbohydrate (NSC) content of *Stipa grandis* and *Leymus chinensis* in 2004 and 2005 (Different superscripts a,b indicate significant ( $P < 0.05$ ) differences between treatments within years and species for mean NSC content; Standard Error of NSC content for *S. grandis*: 1.3 in 2004 and 1.5 in 2005, for *L. chinensis*: 1.5 in 2004 and 2.4 in 2005)

This observation is confirmed by the presented results, as the two investigated species both contain starch and fructan, but differ decisively in the proportion of these carbohydrates. While *S. grandis* predominantly accumulates starch, *L. chinensis* mainly stores fructan. Both reserve carbohydrate fractions were not significantly influenced by defoliation frequency. However, climatic factors had a strong influence on the total NSC content. In 2005 both species showed increased NSC contents, what can be ascribed to the drought. While plant growth is reduced or ceased due to water deficiency, photosynthesis is less sensitive to water stress resulting in NSC accumulation in the plant tissue of grasses (Brown and Blaser, 1970).

## Conclusions

It can be concluded that defoliation had only minor effects on the mean NSC content in the short-term. Climatic factors, as well as species specific differences, are the major effects influencing the NSC content of the two steppes grasses.

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## Fibre fractions of red clover (*Trifolium pratense* L) at different harvests over two seasons

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### Abstract

The objective of this study was to analyse seasonal changes of acid detergent fibre (ADF) and neutral detergent fibre (NDF) in different red clover cultivars. The study was conducted in 2004 and 2005. Two cuts were performed in the first year, in late July (at full flowering) and mid September. There were three cuts in the second year (May 13, June 23 and August 1). ADF and NDF were determined using samples obtained on the day of cutting. For both ADF and NDF, larger differences were found among the cuts than among the cultivars. The highest NDF value was found in the second cut of the second year (417 g kg<sup>-1</sup>). By contrast, the second cut in the first year produced almost 100 g kg<sup>-1</sup> less NDF (329 g kg<sup>-1</sup>). In the present study, the highest ADF value (325 g kg<sup>-1</sup>) was found in the third cut of the second year, while the second cut of the first year produced around 100 g kg<sup>-1</sup> less ADF (239 g kg<sup>-1</sup>).

Keywords: red clover, *Trifolium pratense* L, acid detergent fibre, neutral detergent fibre, forage dry matter

### Introduction

Red clover (*Trifolium pratense* L.) quality depends primarily on the stage of development at cutting, height of cutting and environmental conditions. Taylor and Quesenberry (1996) state that crude protein and *in-vitro* digestible dry matter (IVDDM) are the two most indicative quality parameters. Values of both parameters decrease with age in all perennial forage legumes, as a result of a reduced leaf to stem ratio and lignification. Height of cutting significantly affects forage quality; at budding stage, the upper half of the plant was found to be more digestible than the lower part (Wilman and Altimimi, 1984). Digestibility decline after budding stage is due to increased lignin content and reduced digestibility of non-starch polysaccharides. From the aspect of ruminant rations, the ratio of non-structural and structural carbohydrates plays a special role. The structural carbohydrates are hemicellulose and cellulose whereas the non-structural carbohydrates are starch and sugar. Cell walls of plants consist of hemicellulose, cellulose, lignin, protein and pectin. The cell walls often are determined as neutral detergent fibre (NDF) that contains no pectin because of solubilization of pectin in the neutral detergent solution. In ruminants, the amount of rumination time is directly correlated with the NDF portion in the ration (Grubic and Adamovic, 2003). Treatment of this fraction with an acid detergent helps to separate a fraction called acid detergent fibre (ADF), which consists of lignin, cellulose and some nitrogen. The objective of this study was to assess seasonal changes in fibre fractions (NDF, ADF) of five leading red clover cultivars in the Republic of Serbia.

### Materials and methods

An experiment with five red clover cultivars was established at the end of March 2004 at the experimental field of the Institute of Field and Vegetable Crops in Novi Sad. It included three domestic (K-17, Kolubara, Una) and two foreign cultivars (Viola, Lufa). The plot size was

5 m<sup>2</sup> and the seeding rate was 15 kg ha<sup>-1</sup>. Contents of fibre fractions (NDF, ADF) in the dry matter (DM) of red clover were analysed in two growing seasons (2004 and 2005) for each cut in five replications. The stage of maturity of red clover at each cut was early bloom, except for the first cut in 2004, which was in full blooming. The average annual temperature in Novi Sad is 11.0 °C and the average annual precipitation is 597 mm.

Table 1. Date of cutting and seasonal variation of metrological parameters in 2004 and 2005.

Cut	Date of cutting	Number of days between cuts	Precipitation (mm)	Mean daily temp. °C	Mean max. temp. °C	Mean min. temp. °C
I	21 July 2004	113*	302	18.3	24.2	12.4
II	11 Sep. 2004	52	105	20.6	26.5	14.6
I	13 May 2005	59*	431*	12.0	24.7	-1
II	23 June 2005	41	146	18.3	29.3	8.3
III	01 Aug. 2005	39	129	21.5	33.0	12.0

\*113 – number of days between sowing date and the first cutting.

\*59 – number of days between date of beginning of growing season (15 March 2005) and the first cutting.

\*431 mm – sum of rainfall between 11 Sep. 2004 and 13 May 2005.

Herbage was dried at 60 °C for about 48 hours. Particular attention was paid to making sure that the samples were homogenized and ground to a particle size of 0.8 mm. Chemical analyses were carried out using the filter bag technique (Ankom Technology) for NDF and ADF. All of the analyses were performed on a *Fibre Analyzer Ankom 2001* (Ankom, USA). Finally the hemicellulose content was calculated as the difference between NDF and ADF. The two-factorial analysis of variance was used with cultivar and cut as factors. When the overall *P*-value was < 0.05 the LSD test was used for pair-wise comparisons among means.

## Results and discussion

The results obtained (Table 2) showed that the tested red clover cultivars did not differ significantly with regard to the fractions of fibre in dry matter. There were not any interactions between cultivars and cuts.

Table 2. Variations among red clover cultivars in NDF, ADF and hemicellulose contents (g kg<sup>-1</sup> DM) in 2004 and 2005.

Cultivar	NDF	ADF	Hemicellulose (NDF-ADF)
K-17	375	296	79
Kolubara	374	286	88
Una	361	291	70
Viola	357	284	73
Lufa	379	288	91
SEM	4.2	2.1	4.1
<i>P</i> -value	0.80	0.44	0.62

Significant and expected differences between the analysed fractions of fibre were found among the cuts (Table 3). The NDF values were higher in the second and third cuts of the second year compared with the other cuts, probably related to the rapid ageing caused by high temperatures during summer months. Similar results were obtained by Katic *et al.* (2007) while studying seasonal variations in fibre contents of lucerne in the second year of cultivation.

Taylor and Quesenberry (1996) state that environmentally caused stress and seasonal variations may significantly reduce the quality of red clover forage, as a result of a reduced leaf to stem ratio and lignification. However, a study of Buxton *et al.* (1985) showed that

quality decline with age is not as drastic in red clover as in some other perennial legumes when the leaf to stem portion is considered (0.25 in red clover, 0.16 in birdsfoot trefoil and 0.06 in lucerne). These authors also state that the digestibility of red clover falls by about 8 g kg<sup>-1</sup> node<sup>-1</sup> going from stem tip to base; in lucerne and birdsfoot trefoil, the corresponding reduction is about 20 g kg<sup>-1</sup> node<sup>-1</sup>.

The NDF and ADF concentrations obtained in the second cut of the year of establishment were low but such values were optimal for ruminant rations (Table 3). According to Grubic and Adamovic (2003), rations of highly productive dairy cows in lactation should contain at least 250-280 g kg<sup>-1</sup> of NDF and 190-210 g kg<sup>-1</sup> of ADF. The same authors state that ruminant rations should contain 350-420 g kg<sup>-1</sup> of non-structural carbohydrates. Larger amounts of sugars and readily fermentable carbohydrates may result in acidosis and decline in the proportion of milk fats.

Table 3. Variations in NDF, ADF and hemicellulose contents (g kg<sup>-1</sup> DM) of red clover at different cuts in 2004 and 2005.

Cut	Date of cutting	Phenological phase	NDF	ADF	Hemicellulose (NDF-ADF)
I	21 July 2004	Full bloom	357	294	63
II	11 Sep.2004	Early bloom	329	239	91
I	13 May 2005	Early bloom	352	290	63
II	23 June 2005	Early bloom	417	298	118
III	01 Aug. 2005	Early bloom	392	325	66
SEM			15.4	14.2	10.9
P - value			0.002	0.001	0.01
LSD <sub>0.05</sub>			32	22	35

## Conclusions

The study of the contents of fibre fractions (NDF, ADF) and hemicellulose in the five red clover cultivars tested for two years (2004-2006) showed that statistically significant differences existed among individual cuts but not among cultivars.

Both NDF and ADF concentrations increased during the summer period due to high temperatures and accelerated ageing of plants. Knowledge of seasonal variations in the quality of red clover forage is of special importance for providing a satisfactory proportion of structural and non-structural carbohydrates in ruminant rations.

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## Feeding value of legumes and grasses at different harvest times

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### Abstract

White clover (*Trifolium repens*), red clover (*Trifolium pratense*), lucerne (*Medicago sativa*), perennial ryegrass (*Lolium perenne*) and festulolium (*Festulolium*) plots were established in 2005 and were harvested four times in 2006. In the spring growth and in the second regrowth three maturities were harvested at one-week intervals. Chemical analysis, *in-vitro* digestibilities and *in situ* NDF degradabilities were performed, and they revealed large variation in feed quality between forage types, maturities and especially between the spring growth and the second regrowth. With increased maturity, grasses and lucerne generally showed reduced digestibilities, whereas clover showed negligible reductions except for red clover in the second regrowth. Both grasses and legumes showed considerably lower *in-vitro* organic matter digestibility (IVOMD) in the second regrowth compared to the spring growth. Legumes and grasses differed considerably in relationships between IVOMD and fibre measures as NDF, ADF and ADL, with lower NDF and higher ADL concentrations in legumes compared to grasses at similar IVOMD. Further, at similar IVOMD, proportions of potentially degradable NDF (pdNDF) of total NDF was lower, and the rate of degradation of degradable NDF (dNDF) was higher in legumes compared to grasses. Indigestible NDF (iNDF) to ADL ratio was higher in grasses than in legumes, and the difference increased with increasing ADL concentration.

Keywords: lucerne, white clover, red clover, perennial ryegrass, festulolium, iNDF

### Introduction

An effective milk production is dependent on highly digestible forage, to assure high forage intake and thereby a high forage/concentrate ratio, which is important to maintain a healthy rumen. In Denmark the main forages at present are maize for silage and grass/clover for pasture and/or silage, where perennial ryegrass/white clover swards are common for pasture and perennial ryegrass/red clover swards are common for silage. Festulolium is a new grass in Denmark with a great production potential, and is mainly used to supplement perennial ryegrass in mixed swards for silage. Lucerne is a well known legume, highly appreciated in, for example, the USA, but has had a minor position in Denmark for decades. The present experiment was set up to compare the feed value of white clover, red clover, lucerne, perennial ryegrass and festulolium when harvested at different maturities in the first growth and the second regrowth, and to study whether chemical measures could be used to predict biological values like organic matter digestibility and NDF degradability.

### Material and methods

Plots with white clover (cv. Milo), red clover (cv. Rajah), lucerne (cv. Pondus), perennial ryegrass (cv. Mikado) and festulolium (cv. Perun, *Lolium multiflorum* and *Festuca pratensis*) were established in 2005. Grasses and legumes were fertilized with 240 and 0 kg N ha<sup>-1</sup> yearly, respectively. In 2006 four cuts were harvested, but only the spring growth and the second regrowth were examined. No samples were taken from the first regrowth harvested on 28 June. Plots were harvested 17, 23 and 31 May (red clover was further harvested 7 June) in

the spring growth and 1, 8 and 16 August in the second regrowth. Samples were dried at 60 °C and milled on a 1 mm screen before chemical analysis according to conventional methods and *in-vitro* organic matter digestibility (IVOMD) according to Tilley and Terry (1963). Samples used for *in situ* studies were freeze dried and milled on a 1.5 mm cutter mill. *In situ* studies were conducted according to the NORFOR standard as proposed by Erikson *et al.* (2005), where indigestible NDF (iNDF) and potentially degradable NDF (pdNDF) were estimated using 12 µm pore size bags for 288 h.

## Results and discussion

In the spring growth, IVOMD for white clover and red clover was rather constant over the three harvest times, whereas IVOMD for perennial ryegrass decreased by 0.04 units, and IVOMD for festulolium and lucerne decreased by 0.07 and 0.09 units from first to last harvest time, respectively. In the second regrowth, only IVOMD for white clover was constant over harvest times, whereas IVOMD for perennial ryegrass, festulolium, red clover and lucerne decreased by 0.07, 0.07, 0.07 and 0.05 units, respectively. However, digestibility for the second regrowth was generally much lower than for the spring growth, and IVOMD for perennial ryegrass, festulolium, white clover, red clover and lucerne were 0.14, 0.19, 0.15, 0.10 and 0.19 units lower for the first harvest time in the second regrowth compared to the first harvest time in the spring growth (results not shown). In an earlier article from an accompanying study it was shown that the main reason for decreasing OM digestibility with increased maturity was an increase in the stem/leaf ratio, whereas digestibilities of stem and of leaf were unaffected by maturity (Søgaard and Weisbjerg, 2007).

Some general relationships between measures are shown in Figure 1a-1f.

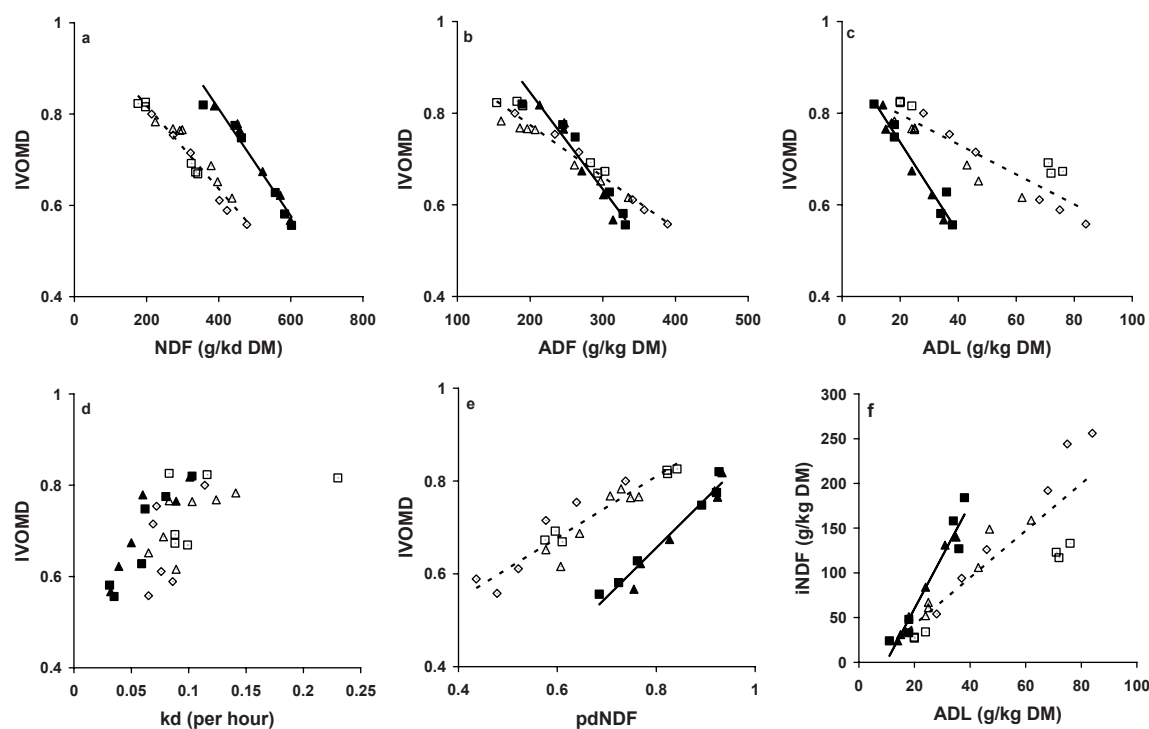


Figure 1. a-e: *In-vitro* organic matter digestibility (IVOMD) plotted against NDF, ADF and ADL in g kg DM<sup>-1</sup>, against rate of degradation of degradable NDF ( $k_d$ , h<sup>-1</sup>) and against potentially degradable NDF (pdNDF). f: Indigestible NDF (iNDF) (g kg DM<sup>-1</sup>) plotted against ADL (g kg DM<sup>-1</sup>). ▲ perennial ryegrass ■ festulolium □ white clover Δ red clover ◇ lucerne. Full line and slatted line are regression lines for grasses and legumes, respectively.



In Figure 1a IVOMD was plotted against NDF concentration in the DM, revealing a high correlation between OM digestibility and NDF concentration within forage type; however, there was a large difference between legumes and grasses.

In Figure 1b IVOMD was plotted against ADF concentration in DM, again revealing a tight correlation. For ADF the difference between legumes and grasses is on average minor; however, the slope is considerably steeper for grasses than for legumes. This seemed mainly to be due to a higher IVOMD in legume stems than in leaves at the same ADF (data not shown). ADF is the chemical component with the highest universal correlation to IVOMD for the whole material.

In Figure 1c IVOMD was plotted against ADL. As for NDF, different relationships were obvious for legumes and grasses with considerably higher lignin content in legumes than in grasses at the same IVOMD. Compared to other legume samples, white clover samples from the second regrowth showed a higher digestibility than expected from their lignin concentration, probably due to the high flower proportion in white clover (around 50% of DM) in the second regrowth (Søgaard and Weisbjerg, 2007). Compared to red clover, lucerne samples showed a higher ADL concentration at similar IVOMD.

In Figure 1d IVOMD was plotted against *in situ*  $k_d$  for dNDF, revealing that at the same digestibility legumes compared to grasses showed a higher rate of dNDF degradation, due to the lower pdNDF in legumes. One sample (second harvest time in spring growth) of white clover showed a very high and unexplainable  $k_d$ .

In Figure 1e IVOMD was plotted against pdNDF, revealing two very distinct relationships between IVOMD and pdNDF, where the same IVOMD was seen for legumes at a much lower pdNDF compared to grasses, due to the lower NDF concentration and the higher rate of dNDF degradation for legumes. Lucerne samples seemed to be more distinct from grasses than white and red clover.

In Figure 1f iNDF concentration was plotted against ADL concentration, revealing that the iNDF/ADL ratios for legumes and grasses were nearly equal at low iNDF and ADL concentrations, but considerably higher for grasses than legumes for samples with high iNDF and ADL concentrations. Further, the white clover samples from the second regrowth were distinct with a very low iNDF/ADL ratio probably due to the high flower proportion.

## Conclusion

Correlations between fibre concentration, composition and degradability differed considerably between grasses and legumes invalidating most universal relationships across both legumes and grasses for predicting OM digestibility and fibre degradability. Within forage type, correlations between biological measures as OM digestibility and NDF degradability and different measures for fibre concentration (NDF, ADF, ADL) were generally high. However, for legumes, flowering white clover deviated from the general legume relationship for IVOMD:ADL and iNDF:ADL ratios. Further, lucerne seemed to deviate slightly from the general legume relationships for the IVOMD:pdNDF ratio. Presented relationships are based on only one year's growth, although on two very different growths each with three maturities, and should be judged in that light.

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## **Session 3**

### **Efficient resource utilisation for sustainable conventional and organic grassland production**



# Sustainable nutrient management for organic farming systems

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## Abstract

In organic farming, soil management largely controls the supply of nutrients to crops, and subsequently to livestock and humans. Crop rotation, including a mixture of leguminous 'fertility building' and cash crops, is the primary method for nitrogen (N) supply within organic systems. Organic rotations are divided into phases that increase the level of soil N and phases that deplete it. In addition to symbiotic N fixation and atmospheric deposition, nutrients may be brought in to the organic system in imported animal feeds, manures, composts and permitted fertilizers. One of the key challenges in supplying N to organically produced crops and forage and preventing gaseous and leaching losses is synchronizing N supply and demand both seasonally and through the rotation. Organic farming favours outdoor livestock production and there is need to develop management systems which minimize nutrient losses from this aspect of production. In order to prevent declines in soil nutrients it is important to balance purchases and sales of phosphorus, potassium and trace elements. The overall nutrient use efficiency of organic systems is governed by the specific cropping and management practices used on each farm.

Keywords: nitrogen, nutrient cycling, organic farming, phosphorus, potassium

## Introduction

Organic farming systems rely on the management of soil organic matter to enhance the chemical, biological, and physical properties of the soil, in order to optimize crop production. Soil management controls the supply of nutrients to crops, and subsequently to livestock and humans. One of the fundamental differences between management of organic and conventional systems is the way in which problems are addressed. Conventional agriculture often relies on targeted short-term solutions, e.g. application of a soluble fertilizer. Organic systems, in contrast, use a strategically different approach, which relies on longer-term solutions (preventative rather than reactive) at the systems level. Organic farming systems are very diverse and are found across the world with a range of crop and animal enterprises often linked together. However, strong unifying principles link this wide range of farming systems and management practices (IFOAM, 2005) and in many countries organic farming now has a clear legislative basis and certification schemes for production and processing. Within the EU, crop and livestock products sold as organic must be certified as such under EC Regulation 2092/91 and 1804/99.

In the European Union (including the new member states) there were 6.9 million ha (3.9% agricultural land area) and 190,000 organic farms at the end of 2005 (Willer and Yussefi, 2007). Organic farming systems fall into similar categories as those of conventional agriculture: mixed, livestock, stockless and horticultural. With the exception of Cyprus and Finland, most European countries have a significant proportion of organic land under grass. Italy which has the biggest fully converted organic area has 23% under grass, the UK has 73% and both the Czech Republic and Slovenia have over 90% (Llorens Abando and Rohner-Thielen, 2007).

Minimizing pollution is high up the agenda for organic farming (IFOAM, 2005). The potential of such systems to provide positive environmental benefits, especially biodiversity, has been demonstrated in a number of countries (e.g. Condrón *et al.*, 2000; Hansen *et al.*, 2001; Hole *et al.*, 2004). Within this paper we explore some of the nutrient management options open to organic farmers, giving examples of their impact on productivity and the environment. We concentrate primarily on forage production but owing to the complex, rotational nature of most organic production we will also refer to farming systems which include arable production. We examine the management of nitrogen (N), phosphorus (P) and potassium (K) in turn and also make some comments on the management of trace elements. A key background issue throughout is, of course, the maintenance and increase of soil organic matter levels due to its key role in nutrient supply, soil structure and water holding capacity. We do not focus on manure management except where there are issues specific to organic production as the principles of good management of manures and wastes apply equally in organic and conventional production.

### **Nutrient management options in organic farming**

The EU regulation on organic farming sets out the main mechanisms for nutrient management (Box 1). Crop rotation, including a mixture of leguminous 'fertility building' and cash crops, is the primary method for N supply within organic systems. Organic rotations are divided into phases that increase the level of soil N and phases that deplete it. The N building and depleting phases must be in balance, or show a slight surplus, if long-term fertility is to be maintained (Berry *et al.*, 2002). This type of rotation provides the basis for forward planning of N supply, necessary in the absence of soluble N fertilizer. The ratio of ley to arable will be determined by a combination of the system (stocked or stockless) and the soil type, being lower on N-retentive soils and higher on sandy soils. In North West Europe, a typical rotation on a mixed organic farm with a three-year grass and clover ley will support two or three years of arable cropping. This may be extended by including an N-fixing cash crop, such as beans, or by including a short period of N-fixing green manure such as vetch between cash crops. Cultivation itself leads to an increase in nutrient availability, particularly N, as microbial activity is stimulated and organic matter breakdown occurs (Silgram and Shepherd, 1999). Mechanical weed control, commonly used in organic horticultural systems, can thus provide a mid-season boost to crops by stimulating mineralization although at other times additional stimulation of mineralization may cause losses by leaching or denitrification.

#### **Box 1. Extract from Council regulation (EEC) No 2092/912.1**

The fertility and the biological activity of the soil must be maintained or increased, in the first instance, by:

- a) cultivation of legumes, green manures or deep-rooting plants in an appropriate multi-annual rotation
- b) incorporation of livestock manure from organic livestock production in accordance with the provisions and within the restrictions of part B, point 7.1 of this annex
- c) incorporation of other organic material, composted or not, from holdings producing according to the rules of this Regulation.

In addition to symbiotic N fixation and atmospheric deposition, nutrients may be brought in to the organic system in imported animal feeds, manures, composts and permitted fertilizers. The

nature and quantity of imported nutrients will depend on the system and the soil type. Some reliance on bought-in feed and bedding on organic dairy farms and purchased manure in organic horticultural systems is shown in Watson *et al.* (2002). Manures from non-organic livestock production may be brought onto the holding but there are restrictions (e.g. it must originate from an 'ethical' source and the animals producing it must not have been fed on a diet containing Genetically Modified Organisms). In order to balance the offtake of specific nutrients there are a number of mineral nutrient sources acceptable in organic systems although their use is permitted only where the need can be demonstrated to the certifying body (for example by soil analysis or by presentation of a nutrient budget). Amendments include rock phosphate, rock potassium, magnesium rock and gypsum. Products such as rock phosphate release nutrients over a period of years rather than weeks (Rajan *et al.*, 1996) and thus their use is planned to build fertility in the longer-term. Trace elements may also be applied, with approval, if they are necessary. The use of lime to maintain pH levels is also acceptable.

### **Nitrogen use in organic farming**

#### *Nitrogen inputs*

Nitrogen is a key factor for crop growth in all farming systems. Biological N-fixation as a result of the legume-rhizobium symbiosis is the main source of N in organic production. Hence, N fixing crops present an important option for improving N supply and maintaining soil fertility (Stockdale *et al.*, 2001). The amount of N derived from N-fixation by legumes depends on the farm structure. In a comparison of nine organic farms, legumes account for 70% of the N supply of crops in stocked systems while in stockless systems a proportion of 42% comes from legumes and 35% from manures (Berry *et al.*, 2003).

#### *Nitrogen budget*

Nitrogen budgets, a core indicator for efficiency and sustainability of nutrient management, usually show a surplus of N for organic farming systems at farm scale. In the study of Berry *et al.* (2003) seven of nine organic farms with significant proportions of arable cropping in the rotation showed positive budgets of 18-64 kg N ha<sup>-1</sup> and two showed negative budgets (-15 and -19 kg N ha<sup>-1</sup>). These positive figures were in line with those of other studies with 30-38 kg ha<sup>-1</sup> (Kaffka and Koepf, 1989; Nolte and Werner, 1994) indicating a long-term N-accumulation in the soil. Hence, N does not seem to be a limiting factor for productivity on organic farms because of the high fixation rates in pastures and clover leys (Nguyen *et al.*, 1995). Although there appeared to be an N surplus, this was not always reflected in grain high N concentrations, probably because of poor synchronization of mineralized N with crop requirements. Furthermore, high N surplus is not always reflected in increased N concentrations in the top soil and thus there is a risk of N loss from this soil layer (Boldrini *et al.*, 2007). Unutilized N can be lost either through leaching or in gaseous forms.

Low N surplus indicates a high N efficiency. In a comparison of an organic all-arable crop rotation with a corresponding mixed farming system, Loges *et al.* (2007) detected significantly higher yields, higher energy efficiencies and lower nitrate leaching in the mixed farming system. Topp *et al.* (2001) calculated a higher N efficiency for rotational cropped land (81%) than for permanent grassland (74%) based on the ratio of N input to saleable outputs for an organic farm in Scotland. The corresponding net change in soil N storage was 22 kg N ha<sup>-1</sup> on rotational land and 38 kg N ha<sup>-1</sup> on permanent grassland.



### *Nitrogen losses*

In organic arable cropping systems a lack of synchronisation of N supply from ploughed leys or grain legumes with the requirements of the following crops is a major problem. While mineralization boosts N contents in the soil shortly after ploughing, N is actually required over the following 3-5 years (Nguyen *et al.*, 1995). Crop yields are often significantly lower in the latter stages of the arable phase (Watson *et al.*, 1999) possibly due to lack of available N. Clearly the variables of time of ploughing and time of cessation of grazing before ploughing are important in determining the rate of production and fate of mineralized N, i.e. as crop uptake or as loss by leaching or as gases (Korsaeth *et al.*, 2002). In a comparison of a mixed system and a pure cropping system, Loges *et al.* (2007) found that the management of grass/clover (mulching versus feeding) had the strongest influence on nitrate leaching in the organic systems. Ball *et al.* (2007) suggest that the optimal time of ploughing for reduced losses as nitrous oxide (N<sub>2</sub>O) is in early spring when the cold temperatures restrict mineralization but N becomes available as the soil warms up and crops require N. Davies *et al.* (2001) suggested that leaving leys ungrazed and unfertilized over winter before ploughing in spring could also reduce the substantial short-term input of N<sub>2</sub>O to the atmosphere that they found after autumn ploughing. Ball *et al.* (2007) also suggest that early cessation of grazing is helpful in that it leaves an adequate supply of narrow C:N ratio residues for incorporation. In a study of farms in five European countries, Petersen *et al.* (2006) used a combination of modelling and measurement to compare losses of nitrous oxide from organic and conventional dairy farms. They found losses were generally lower from organic systems; however, in all cases there was a strong relationship between N input and N<sub>2</sub>O loss.

Even if there is a potential risk of N loss, organic farms often showed similar or lower nitrate leaching losses than conventional ones. Van Diepeningen *et al.* (2005), in a comparison between organically and conventionally managed farms, found significantly lower levels of both nitrate and total soluble N in the organically managed soils. The levels of ammonium and organic N did not differ significantly between the management systems. Nitrate leaching under the same cropping conditions were similar or slightly higher in conventional farms. Stopes *et al.* (2002) also showed similar leaching losses between organic and conventional systems. Using a modelling approach, Topp *et al.* (2000) determined that N losses were lower from the rotational organic area than the conventional one, although for the permanent pasture the losses were similar. At the whole farm level the losses from the organic system were 62% of those calculated for the conventional system.

For animal welfare reasons, organic farming favours outdoor livestock husbandry. Hence, risk evaluation of N loss in organic farming should also address livestock management. The excretory behaviour of pigs, poultry and cattle may create plant nutrient hotspots in outdoor areas, increasing the environmental impact. In an outdoor experiment on pig fattening N concentrations were found to be between 3 to 8-fold higher (reaching levels of 230 to 350 kg ha<sup>-1</sup> in 0-90 cm depth) in defecating areas than in other parts of the pen (16 to 72 kg ha<sup>-1</sup>) (Salomon *et al.*, 2007). In outdoor runs of laying hens ammonia emissions from the pens were relatively small compared to those from the hen house, but the N load within a radius of 20 m of the hen house exceeded a threshold value for N supply from manure (170 kg N ha<sup>-1</sup>) by a factor of 15. Grazing dairy cows gather forage on areas of about 60 to 100 m<sup>2</sup> a day and deposit their faeces on rather small patches of about 3 m<sup>2</sup> size (Haynes and Williams, 1993). The urine-affected areas of cattle grazed grassland swards receive inputs of N at rates equivalent to 300-1300 kg N ha<sup>-1</sup> (Whitehead, 1970). In fact, cattle husbandry plays a dominant role in the nutrient management of organic mixed or specialized farms. It has been suggested that the conversion of specialized dairy farms on sandy soils from highly intensive

conventional practices to organic standards could significantly reduced nitrate inputs into groundwater with only a slight reduction in forage yields (Taube *et al.*, 2006). Even in unfertilized mixed systems, N-fixation by legumes can exceed the amounts of N removed via animal products, resulting in nitrate concentrations in drainage water well above the EU limit for potable water. Nitrate leaching losses in a rotational grazing system on sandy soils could be reduced by the inclusion of one or two silage cuts in spring. In terms of leaching losses, a cutting-only system is very advantageous due to high N export rates (Wachendorf *et al.*, 2004).

### **Phosphorus and potassium use in organic farming**

It has been suggested that organic farmers mine soil supplies of P and K built up by historic fertilizer applications (e.g. Johnston *et al.*, 1991; Greenland, 2000). There is some evidence in the literature of reduced levels of available P and K in soils on organic farms compared to conventional (Gosling and Shepherd, 2005). Loes and Ogaard (1997) found reductions in available P and K in soils with previously high levels following conversion to organic farming as a result of reduced imports. In contrast, Kaffka and Koepf (1989), who studied a biodynamic dairy farm that had been in operation since 1929, found only small differences in soil P and K content over time. Manures are clearly a valuable source of P and K whether they are produced within the farm system or imported. Fewer published values of P and K are available for manures from organic farms than from conventional farms, but data from both Steineck *et al.* (2000) and Shepherd *et al.* (2002) suggest that nutrient concentrations are generally lower in manures from organic farms compared with conventional. As manure is such an important source of P and K in organic systems, manure needs to be used tactically such that manure is applied to grassland cut for silage rather than during the grazing period when P and K are being deposited in dung and urine. A range of additional P and K sources can be imported under current organic farming standards, these are characterized as being of low solubility and being made available through soil processes rather than directly to the plants. Some of the fertilizers can be applied at the discretion of the farmer, e.g. rock phosphate, while others require permission from the certifying body e.g. potassium sulphate.

### *Phosphorus cycling*

The key role that crop choice and rotation holds in managing nutrients was set out in Box 1 and has been discussed above for N. It is also critical for effective use of P. In organic farming systems, where the use of soluble P fertilizer is prohibited, it has been suggested that P deficiencies might critically limit rotational yields because of the impact of P deficiency in reducing the N<sub>2</sub> fixation capacity of legumes (Romer and Lehne, 2004) It is therefore important to consider other agronomic approaches that might be used to improve the effectiveness of P use within the farming system. Many plants are able to respond to P shortage and may increase P availability by: adaptation of root system morphology (increasing root surface area and/or root hair development); changing soil pH or excreting chelates in the rhizosphere to solubilize inorganic forms of P; releasing hydrolytic enzymes which enable soil organic P to be used by the plant, and/or association with mycorrhizal fungi. These responses have a direct benefit for the crop itself, but may also have benefits for the crop rotation as a whole, if P is mobilized from recalcitrant soil P fractions and mobilized P is made available to following or adjacent crops. Crop rotation design can also have an impact on P availability indirectly through the modification of soil properties – particularly soil structure and microbial activity (Horst *et al.*, 2001). Cultivation (McGonigle and Miller, 2000) and the inclusion of non-mycorrhizal crops (e.g. brassicas) within the rotation (Karasawa *et al.*, 2001) can reduce survival and effectivity of arbuscular mycorrhizal fungi. Designing crop

rotations for increased P efficiency in organic farming thus means the targeted inclusion of crops and cultivars with high P uptake efficiency particularly as inter-crops or as cover crops. For example, *Lupinus albus* is well known to develop cluster (proteoid) roots which are able to mobilize sparingly soluble soil P (Braum and Helke, 1994). Other leguminous crops are also able to mobilize soil P through rhizosphere acidification (Vance *et al.*, 2003) and it has been shown that the rotational benefit of legumes is not simply a result of the N fixed. Cavigelli and Thien (2003) showed that the P uptake of the previous crop (for perennial forages) and the plant type (for winter cover crops) influenced the P uptake of a subsequent sorghum crop. In other words, that cover crops/green manures are useful in improving P use efficiency although they are traditionally thought of as being used for N management.

Permitted inputs include rock phosphate and calcined aluminium rock phosphate (for use when pH exceeds 7.5). There is currently much interest in methods for increasing the accessibility of P added in rock phosphate. These include adding rock phosphate to animal manures but also composting or fermenting rock phosphate with green wastes (Stockdale *et al.*, 2006).

Table 1. Farm-scale P budgets by organic farm type, from Watson *et al.* (2002).

Farm type	Surplus (Input-Output) kg ha <sup>-1</sup> yr <sup>-1</sup>			
	n	Mean	s.e.	Range
Arable	1	-6.0		
Beef	4	-1.8	1.4	-6-0
Dairy	56	3.1	0.9	-6.6-36.0
Horticulture	3	38.9	26.0	1.7-89.0
Mixed	6	-2.4	1.3	-6.9-4.0
Mean		3.6		

### *Phosphorus losses*

In the majority of soils, losses of P are most likely to occur through the transport of P associated with colloidal clay or organic matter in both surface run-off and in drainage. These losses are not strongly related to P inputs or the P budget for the system (Edwards and Withers, 1998) and are very closely related to the movement of sediments. Heathwaite *et al.* (1998) showed that greater quantities of P were lost in surface runoff from grassland receiving soluble P fertilizers than from farmyard manure or slurry treatments. Losses of fertilizer through runoff are less likely to occur in organic agriculture where fertilizers are applied less frequently and only relatively insoluble materials (phosphate rock) are used. Leaching of dissolved P will only occur on soils with high P concentrations, likely to be the result of long-term over-fertilization and/or excessive applications of animal manures where the accumulation of P exceeds the soils sorption capacity (Haygarth and Jarvis, 1999). As P budgets for organic farms rarely show a significant surplus of P (Table 1) losses by this route would be expected to be small.

### *Potassium cycling*

Potassium is potentially the most difficult major nutrient to manage in organic systems since K sold in produce must be replaced, but there is no obvious sustainable source of K available to organic farmers. On some soils, e.g. heavy clays, the release of K from non-exchangeable sources will support crop production without imports (Goulding and Loveland, 1986). In other cases, particularly where high K demanding crops such as forage and potatoes are grown this is not the case. Kayser and Isselstein (2005) quote figures of 3-5 kg ha<sup>-1</sup> yr<sup>-1</sup> for K weathering in coarse sandy soils in N Europe compared with 65-80 from clay rich soils. Where deficiency can be demonstrated organic certification bodies will allow the use of some

materials such as sulphate of potash, MSL-K (volcanic tuff) and Kali vinasse (by-product of the sugar beet industry). There is a need for information on the long and short term effects of newly available materials on soil K status such as the latter two products. Pot experiments and small plot experiments carried out on organic land are entirely appropriate for this type of research. However, as shown by Fortune *et al.* (2004), it is important to look at all the nutrients applied in the added materials and their interactions. Yield responses associated with many of these materials are small, particularly in situations where N is limited (Table 2).

Table 2. Yield of grass/clover (g dm pot<sup>-1</sup>) in a pot experiment (sum of 4 cuts) with a range of organically acceptable K sources applied at 41.5 kg ha<sup>-1</sup> +/- additional N (Fortune *et al.*, 2004).

Treatment	Yield grass/clover (g pot <sup>-1</sup> )	
	-N	+N
Control	7.24	18.85
DKSI	10.46	21.63
Kali vinasse	8.97	17.2
MSL-K	8.29	19.17
Rapemeal	15.69	24.29
Sulphate of potash	9.4	26.35
Sylvinite	8.1	20.54
Farmyard manure	15.92	
MSL-K + FYM	13.09	

Kristensen and Halberg (1995) found K surpluses of 33 and 82 kg ha<sup>-1</sup>yr<sup>-1</sup> in a sample of 17 organic and 19 conventional dairy farms respectively. They found that the K surplus increased with increasing livestock density because of increased feed imports. On the 67 organic farms used by Watson *et al.* (2002) to calculate K budgets, the budgets showed both surpluses and deficits (average 14.2 kg K ha<sup>-1</sup> year<sup>-1</sup>) with horticultural systems showing large surpluses resulting from purchased manure (Table 3). They note the same relationship as Kristensen and Halberg (1995) with respect to imports. Watson *et al.* (2002) also note the need to analyse materials for the calculation of accurate potassium budgets as organic produce may have lower K contents than equivalent conventional produce. As with the other nutrients, farm scale budgets can hide large variations in field budgets, potatoes and silage having particularly large K offtakes.

Table 3. Farm-scale K budgets by organic farm type, from Watson *et al.* (2002).

Farm type	n	Surplus (Input-Output) kg ha <sup>-1</sup> year <sup>-1</sup>		
		Mean	SE	Range
Arable	1	57.0		
Beef	4	3.0	3.4	-4.5 - +12.0
Dairy	58	9.6	2.0	-26.5 - +58.0
Horticulture	3	122.0	88.0	-23.0 - +281.0
Mixed	3	-2.2	1.2	-4.4 - -0.3
Mean		14.2		

Significant K losses can occur from manure heaps, from the disposal of silage effluent and on some soils via leaching. Younie *et al.* (1998) demonstrated the value of silage effluent as a potassium source in organic grassland cut for silage. Potassium loss by leaching is strongly correlated with soil type. As clay content increases, soils tend to retain more K (Askegaard *et al.*, 2004). However, Williams *et al.* (1990) demonstrated that even on K soils, leaching may occur through preferential macropore flow where high concentrations of K occur in urine patches. Potassium losses from grassland have been reviewed recently by Kayser and Isselstein (2005). Potassium in waste materials is as available as mineral forms of K and is



therefore at risk of leaching from uncovered manures and composts. As outlined in Box 1, the guidelines for using crops with different root structures is important in relation to potassium since the use of crops with deep rooting systems within a rotation may allow some K to be recovered from the subsoil. There is also evidence that different varieties of crops, e.g. barley, differ in their ability to exploit soil K (e.g. Siddiqi and Glass, 1983). This suggests that it might be important to screen varieties for this property for organic farming. The inclusion of deep rooting herbs within leys can influence P, K and trace element contents of forage (Whitehead, 2000).

## Conclusions

In conclusion, understanding and improving the efficiency of nutrient cycling in organic systems is fundamental for both maintaining productivity and minimizing adverse environmental impacts. While organic farming systems have the potential to deliver significant benefits for environmental policy, e.g. the Water Framework Directive, the actual effects of changing to organic farming will depend on specific cropping and management practices on each farm. Many of the nutrient management practices used in organic farming are relevant to conventional systems, particularly in these times of increasing fertilizer prices. While we have concentrated on N, P and K in this paper, the management of trace elements is also important for livestock and human nutrition. A major new FORMAS-funded project has been initiated between the UK and Sweden to investigate trace element cycling in soils and forages.

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# Recent developments in harvesting and conservation technology for feed and biomass production of perennial forage crops

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## Abstract

The main objectives were to (a) review recent developments in harvest and ensiling technologies used in Europe, (b) compare the effects of different ensiling methods on physical, chemical and health characteristics and on fermentation profiles of silages, (c) identify further research and development that is required, and (d) review information on biomass production based on perennial grasses. Forage harvesting technologies (mowers, conditioning, spreading and windrowing wilting systems, harvester type) have a key role in energy efficiency, silage costs, nutrient losses and quality of silages. Different ensiling methods (conventional system in bunker silo, tubular system, baled silage using new generation balers, wrapping technology and film types) influence the physical characteristics (homogeneity, chop size, density, anaerobic conditions), fermentation profile (pH, organic acids), nutrient and energy content, aerobic stability and anti-quality features (undesirable microorganisms and chemicals such as mycotoxins) of silages. Harvest management practices of perennial grasses are considered with regard to potential biomass energy feedstocks. A shift towards biomass feedstock production of perennial grasses would enhance soil organic carbon, soil quality, water quality and wildlife habitat, and help revitalize rural economies.

Keywords: silage, harvest, bale, ensiling, biomass

## Introduction

Cool- and warm-season perennial grasses are important sources of feed for livestock. They are also receiving increased attention as sources of renewable energy. The objectives of this paper are to: (a) review recent developments in harvesting and ensiling forage crops in Europe, (b) compare the effects of different ensiling methods on physical, chemical and fermentation characteristics of silages, (c) discuss the need for further silage research and development, and (d) explore biomass production potential of perennial grasses. Forage harvesting technology plays a key role in energy efficiency, production costs, nutrient losses and the quality of silages. Ensiling methods influence physical and quality characteristics of silage as well. Harvest management practices are also being evaluated because perennial grasses are considered as potential biomass energy feedstocks. A shift toward biomass feedstock production of perennial grasses would enhance soil organic carbon, soil quality, water quality and wildlife habitat, help revitalize rural economies and reduce dependence on finite fossil fuel resources.

## 1. Comparison and effect of different harvest technologies

### 1.1. Mowing/conditioning

The type of equipment used to mow and condition forages affects drying rate and potentially the quality of the final product. Mounted, trailed and self-propelled rotary mowers (drum or disk systems) equipped with different conditioning systems (tines or corded rollers) and with a working width of up to 14 m are available to accelerate the wilting process. Conditioning rollers are particularly helpful in increasing drying rate of legumes such as alfalfa (*Medicago sativa*) (Hintz *et al.*, 1999). The close-coupled corded roller system is recommended for conditioning alfalfa in order to reduce leaf loss (Bellus and Kelemen, 2001a, b, c, d). There is an option to apply swinging, fixed or controlled tine conditioners (stronger mechanical impact) in order to accelerate the wilting process in grass since grass leaves are more resistant to shattering (Hajdú and Kővári, 1983). Mowing height will also affect quality as stems contribute more to total biomass as cutting height is lowered. However, a lower cutting height increases soil contamination, with a consequent proliferation of harmful microorganisms (*Escherichia coli*, *Clostridia*, yeast) in the silage. For example, lifting the mowing height from 20 to 100 mm decreased the *Clostridia* proliferation from 84,000 to 4,000 CFU g<sup>-1</sup> in grass silage.

Many forage species should be placed in wide windrows to increase surface exposure to sunshine and thereby reduce drying time (Forristal and O'Kiely, 2005). Spreading systems can speed up the crop drying process (Bellus and Kelemen, 2001c,d), achieving satisfactory DM concentrations within 32 hours throughout the harvesting season provided it does not rain (Frost, 1988). However, during the summer (July-August: the average ambient temperature is between 19-22 °C) in the dry continental zone 4-5 hours are sufficient to reach 350-400 g DM kg<sup>-1</sup> in alfalfa (Bellus and Kelemen, 2001c, d).

Trials in Hungary demonstrated the effects of different conditioning systems and windrow spreading on the drying rate of alfalfa (Bellus and Kelemen, 2001a, b, c, d). Spreading accelerated wilting; however, protein losses were 7 to 20% higher when alfalfa was mechanically spread despite the fact that it was carefully done (Orosz and Sasi, 2006) (Table 1).

Table 1. Drying rate (g moisture decrease kg<sup>-1</sup> forage hour<sup>-1</sup>) and crude protein loss in alfalfa with spreading (carpet windrow) and without spreading (32 hours wilting period) (Orosz and Sasi, 2006).

	Without spreading g moisture loss kg <sup>-1</sup> fresh material hour <sup>-1</sup>	With spreading g moisture loss kg <sup>-1</sup> fresh material hour <sup>-1</sup>
Rotary mower without conditioner	6.2	8.3
Rotary mower with swinging tine conditioner	8.3	13.6
Rotary mower with corded roller conditioner	8.9	11.9

### *1.2. Effect of rainfall and wilting on fermentation process, digestibility and feed intake*

In humid climates, perennial grasses may be ensiled directly or after minimal wilting in the field, especially during wet weather conditions. However, wilting successfully reduces dry matter losses (Mayne and O'Kiely, 2005) and improves silage quality compared to unwilted or poorly preserved silage. Under adverse weather conditions wilting may increase losses (Mayne and O'Kiely, 2005) and has little beneficial effect when fresh forages are treated with formic acid before ensiling. There is always a cost in delaying harvest to avoid wet weather because of increased plant maturity or age. Coblenz and Jennings (2004) evaluated losses of DM and changes in nutritive value for wilting legumes, orchardgrass, bermudagrass and tall fescue. From 0 to 76.2 mm of simulated rainfall were applied to orchardgrass and bermudagrass in 12.7 mm increments at varying times and DM concentrations after cutting. Tall fescue was baled at 24.6% moisture after a 22.9 mm rainfall event and at 9.3% moisture following three rainfall events totalling 71.1 mm. Dry matter losses for orchardgrass and bermudagrass were low (< 2%) if rainfall occurred when the forage moisture content was high, but increased substantially if rainfall occurred when the forage was dry. Dry matter losses were lower in bermudagrass than orchardgrass when rain was applied to each species at hay moisture contents. These differences can largely be explained on the basis of the sugar content of each grass. Perennial cool-season grasses, such as orchardgrass, have much higher concentrations of water-soluble plant sugars than bermudagrass or other warm-season perennial grasses. Therefore, orchardgrass has the potential for more DM loss through leaching.

Leaf shatter was a greater source of DM loss in legumes than in grasses. Rain damage increased with the amount of rainfall and the duration of the rainfall event regardless of species, however. More water was retained by the forage when rainfall intensity was low. Concentrations of fibre components (NDF and ADF) increased, as soluble plant sugars were leached away during the application of simulated rainfall. In addition, the leaching of soluble sugars and other compounds from the forage reduced the energy density and digestibility of these forages as much as 8.9%. In a pattern that is similar to that observed for DM loss, changes in nutritive value were disproportionately large with the first 25.4 mm of rainfall.

Rain damage to wilting forages is more severe when the forage is relatively dry because the plant cells lose their ability to control the passage of soluble compounds in or out of the cell. Fibre components (NDF, ADF and lignin) are not water soluble and remain stable during rainfall events. Their concentrations increase indirectly as sugars are preferentially leached away. This results in decreased nutritive value, daily voluntary intake, digestibility and energy density.

Wilting improves lactic acid fermentation, but can reduce aerobic stability in perennial ryegrasses (O'Kiely *et al.*, 2005). Considerable carotene losses caused by UV-radiation during a long wilting period (48 hours) were found by Orosz and Bellus (2007) in a recent trial with alfalfa. Effluent can be reduced not just by wilting grass (35% DM), but also by the application of different hygroscopic by-products (Szűcs *et al.*, 2001). It was shown that DM-intake was higher with wilted compared to fresh-cut silages, while there were minor or even negative effects on milk yields (Bertilsson, 1987). Digestibility and utilisation in wilted silage may be lowered through contamination with ash (Bertilsson, 1990) and by increased passage rate (Tuori *et al.*, 1993).

### 1.3. Conventional harvest

The pick up wagon with a cutter system and mechanical output transfer equipment can be used in silage making and involves two separate functions: crop pick-up/chop and transport. Popularity of the pick-up wagon system has been in decline because contractors preferred a continuous harvesting system (Forristal and O'Kiely, 2005). However, self-propelled forage chopper-harvesters with high-speed chopping and largely pneumatic grass delivery systems are not energy efficient (Tremblay et al, 1991).

In a recent trial, Frost and Binnie (2005) compared the performance of a self-propelled forage harvester system (SPFH) and a self-loading forage wagon (SLFW) system. The herbage harvested was 23.4 t ha<sup>-1</sup> and 286 g DM kg<sup>-1</sup>. Particle size distribution in the range of 21-80 mm was similar for both systems (66.6% for SPFH and 66.2% for SLFW). Particles in the 0-20 mm category were greater in SPFH harvested herbage (22.1 vs. 6.6%), while particles > 81 mm were higher in herbage harvested by the SLFW (27.3 vs. 11.3%). It was shown that there is significant potential for SLFW silage harvesting systems to maximize output per person (24.8 vs. 12.4 Mg forage hours<sup>-1</sup> person<sup>-1</sup>, respectively) and improve fuel efficiency (0.67 vs. 1.32 litre fuel Mg<sup>-1</sup> forage, respectively).

Arvidsson and Lingvall (2005) compared two types of silage trailers (feed rotor with knives and precision chop system). Herbage (35% DM) harvested by the trailer equipped with a rotor was longer (30% < 40 mm) compared to that harvested by the trailer with a precision chopper (84% < 40 mm). There were no differences in silage density, silage quality or amount of spoiled silage; however, there was a low density in both silages (with high levels of butyric acid). The rotor trailer had a higher loading rate (13,559 kg vs. 8,061 kg forage load<sup>-1</sup>, respectively) and required less energy (0.82 vs. 1.2 l MJ, respectively).

In the case of alfalfa and maize silages, the self-propelled forage chopper-harvester with high-speed chopping and pneumatic forage delivery systems probably will continue to dominate in Europe due to its high output, continuous harvesting method, smaller theoretical chop size and homogenous particle distribution. The potential efficiency of a self-propelled chopper-harvester (3.0-4.2 m wide pick-up adapter and 8-10 km h<sup>-1</sup> speed) in alfalfa (15-17 mm chop length and 30-40% dry matter content) is 200 Mg h<sup>-1</sup> at 80% engine load with 0.5-0.6 kg Mg<sup>-1</sup> fuel consumption, in Hungary (Jován and Kelemen, 1990).

## 2. Comparison and effect of different ensiling methods

Various ensiling methods are practised, but can generally be broadly classified as baled silage, tubed silage, and bunker/piled silage. The type of ensiling method used, along with other factors, affects silage physical characteristics (e.g. homogeneity, particle size and density), fermentation profile (e.g. pH, organic acids), nutrient and energy content, aerobic stability, and anti-quality factors in the form of undesirable microorganisms and chemicals. Characteristics of typical conventional round bale grass silage from Ireland (Forristal and O'Kiely, 2005), round bales made from 'chopped' alfalfa as a new type of baled silage (Orosz and Bellus, 2007 unpublished) and a tubular system (Csermely *et al.*, 2002) are shown in Table 2.

Table 2. Characteristics of conventional round<sup>1</sup> bales (grass) (Forristal and O’Kiely, 2005), new type of round bales (alfalfa) (Orosz and Bellus, 2007 unpublished) and tubular system silage (Csermely *et al.*, 2002).

	Conventional baled silage (grass-Ireland)		New type of baled silage (alfalfa)	
	Typical	Range	290 g kg <sup>-1</sup> DM	520 g kg <sup>-1</sup> DM
Nominal size (m)	1.25 x 1.25		1.13 x 1.22	1.13 x 1.22
Weight (kg)	650	350-1000	904±25.1	657±13
Bale weight (kg DM)	195	-	262±7.3	342±6.6
Coefficient of variation	-	-	1.4%	1.5%
Particle size (mm)			20-40	20-40
Dry matter content (g kg <sup>-1</sup> )	300	160-700	290	520
Porosity (% spore space)	60	50-80	-	-
Wet density (kg m <sup>-3</sup> )			734±10.3	534±7.9
Dry density (kg m <sup>-3</sup> )	130	90-200	213±3.0	278±4.1
Pressurization (bar)	-	-	150	150
Film width (mm)	750	250 <sup>2</sup> -750	750	750
Film thickness (µm)	25	12-30	25-35	25-35
Pre-stretch on wrapper (%)	70	25-70	60	60
No. of layers	4	4-8	6-7	6-7
Film weight (g kg <sup>-1</sup> DM)	4.3			

	Tubular system silage	
	Typical	Range
Nominal size (m)	d = 2,4-3,0 60 m length	d = 2,4-3,6 45-90
Weight (kg)	160-240 x 10 <sup>3</sup>	160-450 x 10 <sup>3</sup>
Bale weight (kg DM)	-	-
Coefficient of variation	-	-
Particle size (mm)	25	20-40
Dry matter content (g kg <sup>-1</sup> )	350-400	300-430
Porosity (% spore space)	-	-
Wet density (kg m <sup>-3</sup> )	580	-
Dry density (kg m <sup>-3</sup> )	174-232	-
Pressurization (bar)	50-60	
Film width (mm)	-	-
Film thickness (µm)	250	230-280
Pre-stretch on wrapper (%)	-	-
No. of layers	1	1
Film weight (g kg <sup>-1</sup> DM)	0.21-0.31	

<sup>1</sup>Various size of large rectangular (e.g. 0.8 m x 0.8 m x 1.2m) and small rectangular bales are also wrapped.

<sup>2</sup>250 mm only used on small rectangular bale wrappers.

## 2.1. Baled silage

### 2.1.1. Conventional bales

Baled silage occurs on a higher proportion of farms than all other systems in humid, temperate areas. In Ireland baled silage, precision-chopped silage, and single/double crop silage are made on 73, 32 and 9% of farms, respectively (O’Kiely *et al.*, 2002). Baled silage typically has a higher pH, a more restricted fermentation, especially at higher dry matter contents (Jones and Fychan, 2002; McEniry *et al.*, 2005), and higher porosity (O’Kiely *et al.*, 2002; Forristal and O’Kiely, 2005) than chopped silage, but the feeding value of the silage is usually similar (O’Kiely *et al.*, 2002). O’Kiely *et al.* (2002) showed individual treatment



effects of bale dry matter concentration, bale density and number of layers of plastic films on the conservation characteristics of bales.

Benefits of baled silage include no silo structural requirement, hay-making equipment may be used for harvest, harvesting energy is reduced by about one-third, and it can be self-fed if properly presented to livestock. However, conditions are not always optimal for fermentation, extreme care must be taken to eliminate air infiltration, it requires prompt wrapping, handling and storage of bales, equipment for lifting and moving bales must be available, and plastic must be purchased but is easily damaged (Bellus and Kelemen, 2001a,b; Orosz *et al.*, 2001).

### 2.1.2. A new baling system

Data for grass are limited; therefore description of this ensiling method is based mainly on alfalfa. Owing to the benefits of this new bale-forming system (Orosz and Bellus, 2007 unpublished), it should be considered as a potentially useful system for grass silage. Terminology of the new system is still being developed.

The crop for bale-forming in the new round baler can be precisely chopped, so a self-propelled chopper-harvester is also required. It is not yet known if a pick-up wagon could be used. The harvest technology could be the same as in the case of clamp silage. Forage can be grass or alfalfa with a theoretical chop length of 20-40 mm depending on dry matter content (300-500 g DM kg<sup>-1</sup>) and fibre concentration (350-400 g NDF kg<sup>-1</sup> DM). There is better homogeneity of the new bales compared to conventional bales, owing to the forage being chopped and mixed (Orosz and Bellus, 2007 unpublished). A high density (213-278 kg DM m<sup>-3</sup>) can be achieved due to high pressurization (150 bar), as well as a short particle size. In one trial, extremely low variation was shown in density among the bales (1.4-1.5% coefficient of variation). High density results in good anaerobic conditions for fermentation. Output and efficiency are as follows: 18-20 bales hour<sup>-1</sup> for alfalfa (Orosz and Bellus, 2007 unpublished). The likely lowest dry matter content in alfalfa new-type bales is 290 g DM kg<sup>-1</sup> (20-30 mm theoretical chop size, 150 bar pressure and 380 g NDF kg<sup>-1</sup> DM) with negligible effluent output. The ideal weight of alfalfa bales (1.1 x 1.2 m size) are 750-800 kg (350-400 g DM kg<sup>-1</sup>, 20-30 mm chop length, 400-450 g DM kg<sup>-1</sup> NDF). For bales heavier than 900 kg there is high risk of effluent production and bale loss (Orosz and Bellus, 2007 unpublished). Combination baling-wrapping machines are available.

### 2.1.3. Bale wrapping

Bale wrapping systems may greatly affect the extent of fermentation and of bale deterioration; therefore, one must consider the limitations of this system. The surface area in contact with film in baled silage is typically 6-8 times that of conventional clamp silage (O'Kiely *et al.*, 2002). Additionally, the thickness of the stretched film used to wrap the bale silage (70 µm for 4 layers) is thinner than double sheeted clamp silos (250 µm for 2 layers) which may create a more aerobic environment at the bale surface (O'Kiely *et al.*, 2002). McEniry *et al.* (2005) found that bales with a mean dry matter concentration of 354 g kg<sup>-1</sup> and pH of 4.5 contained more yeast, *Lactobacillus* and *Enterobacteria* in the outer 20 cm compared to the bale core while there was no significant difference in *Bacillus* and *Clostridia* numbers. Higher yeast and *Enterobacteria* numbers may reflect more aerobic conditions at the bale surface.

Thickness of the plastic film is generally 25 or 35 µm, but there are also films of 12 µm. In a trial with 5 different films, considerable variation in CO<sub>2</sub>-permeation and mechanical

properties was noted across the width of the roll for all polyethylene films, indicating non-perfect manufacture and the need for improved film quality (Laffin *et al.*, 2005).

Mould proliferation is strongly dependent on the number of plastic layers applied. Increasing the number of layers of plastic from four to eight will reduce mould and increase CO<sub>2</sub> level on the bale surface (Lingvall, 1995; Forristal *et al.*, 1999; Heikkila *et al.*, 2002; Jacobsson *et al.*, 2002; O'Kiely *et al.*, 2002; Harrison *et al.*, 2004). Forristal and O'Kiely (2005) established that a minimum of four layers of film is required for effective mould inhibition in temperate climates. A new type of film with low O<sub>2</sub> permeability (triple co-extruded film with two outer layers of polyethylene and a central layer of polyamide in 25 µm thickness) was compared to a commercial polyethylene film (6 layers of 25 µm film). Yeast, mould, and clostridia spores were higher in the bales covered by standard film after 4 months, especially on the surface area. However, there was no significant difference in yeast, mould and clostridia spores between 2 and 4 months on the surface of bales covered with the new type film (Borreani and Tabacco, 2005).

Round bales of grass silage have traditionally been transported un-wrapped from the field to the storage place before wrapping. Other systems (combined baler and wrapper machine) have been developed to allow wrapping in the field before transporting to the storage areas. However, of five systems tested, Randby and Fyhri (2005) found that transport of bales to the storage place (300 m) to wrap (6 layers, 25 µm white plastic film) had given lower mould proliferation than transport of wrapped bales. Bales should be wrapped within 3 hours of baling to minimize DM and quality losses (Kelemen and Bellus, 2001a, b). In the US, Under-sander *et al.* (2005) found that rectangular bales wrapped more than 24 hours after baling had significantly higher neutral detergent fibre than bales wrapped sooner, indicating loss of non fibrous carbohydrate due to respiration.

A combined baler/wrapper machine is recommended only for grass, but should not be used with alfalfa because of definite damage on the plastic foil by the hard, perforating stubble (Bellus and Kelemen, 2001a, b, c).

O'Kiely *et al.* (2002) showed the influence of film colour on silage preservation and mould growth on big bale silage: film colour did not influence any of the measured silage or mould parameters significantly in a temperate climate (Ireland).

Film damage can be caused by inappropriate management, machinery, stubble and wildlife (bird, cat, fox, mouse, rat). O'Kiely *et al.* (2002) evaluated different methods to prevent wildlife damage on big bales. The most effective prevention against wildlife was to move the bales into a safe storage place before wrapping. Use of a net could be effective against bird attack. Chemical repellent should be applied against rodents.

Plastic film can be considered an environmental challenge. Film used for covering round bales is approximately 23.5 kg ha<sup>-1</sup>, for sealing conventional bunker silos is 4.7 kg ha<sup>-1</sup> in the case of grass silage (Forristal and O'Kiely, 2005) and for the tubular system is 2.6 kg ha<sup>-1</sup> for alfalfa silage (Csermely *et al.*, 2002). However, there is an option to collect and recycle the film and there are experiments to develop biodegradable polyethylene (O'Kiely *et al.*, 2002; Forristal and O'Kiely, 2005).



## 2.2. Tube silage

Tube silos are suitable for both large (> 100 dairy cows) and small (minimum of 25 dairy cows) farms. Silage crops are harvested by conventional equipment or a self-propelled chopper-harvester and packed in a tube 30-100 m long and 2.4-3.7 m in diameter. This is a relatively fast ensiling method with an average rate for alfalfa of 100-120 Mg hour<sup>-1</sup> in a 3 m x 60 m tube with continuous service. In practice the general rate is 1 silage tube filled with alfalfa per 8 hours (56 m productive length). Achieving and maintaining anaerobic conditions is rather easy, but punctures in the bag must be avoided to minimize aerobic deterioration (plastic tube: 230-280 µm thickness; 3 layers: UV stable outer layer). A high density of 174-232 DM kg m<sup>-3</sup> can be achieved (Csermely *et al.*, 2002) due to high pressurization and small chop length. The theoretical chop length for alfalfa silage is 20-40 mm (depending on dry matter content: 300-430 g DM kg<sup>-1</sup> and fibre concentration: 350-400 g NDF kg<sup>-1</sup> DM) and in maize silage is 5-20 mm. Higher homogeneity can be achieved compared to baled silage. Anaerobic conditions strongly depend on management. The environmental load of plastic film (2.6 kg ha<sup>-1</sup> alfalfa) is lower than in the baled silage system. Farmers often have a problem with unloading this silage, with consequent impacts on soil contamination (depending on surface: concrete, macadam or soil) and animal health.

Over two years, 47 bag silos (polyethylene tubes, 30 to 90 m length, 2.4 to 3.7 m diameter, 220 µm thick) were investigated in a trial in the US (Muck and Holmes, 2005). Density ranged from 160 to 280 kg DM m<sup>-3</sup> in alfalfa and whole-crop maize and decreased as DM content and particle length increased. Dry matter losses were measured in 39 of the bag tubes and ranged from 0 to 40% with an average DM loss of 9.2% invisible plus uncollected losses and 5.4% spoilage losses for a total loss of 14.6%.

There is a mechanical compression treatment by the plastic rotor which affects the structure of the plant material during the filling of the plastic tube. This function requires considerable energy, and may facilitate effluent release. On the other hand, Sundberg and Pauly (2005) confirmed that the mechanical treatment of forage by the packing rotor had a beneficial effect on the rate of pH decline and on organic acid composition. For grass prior to and after passage through the rotor the pH on day 3 of ensilage was 4.85 and 4.41, respectively, with corresponding lactic acid values of 36 and 45 g kg<sup>-1</sup> DM.

Dry matter losses can be minimized by ensiling at the proper moisture concentration (350-400 g DM kg<sup>-1</sup> for alfalfa in a dry climate) (Csermely *et al.*, 2002). This will also help alleviate compaction problems and better maintain silage quality (Orosz *et al.*, 2001). Schmidt *et al.* (2002) also demonstrated the effect of dry matter concentration on quality characteristics of tubed alfalfa silage (Tables 4 and 5).

Table 4. Effect of different dry matter contents (short-term wilting) in alfalfa ensiled in plastic tubes (Schmidt *et al.*, 2001).

	pH		Lactic acid		Acetic acid		n-butyric acid		NH <sub>3</sub>	
			g kg <sup>-1</sup> DM		g kg <sup>-1</sup> DM		g kg <sup>-1</sup> DM		g kg <sup>-1</sup> DM	
Day of fermentation:	20	60	20	60	20	60	20	60	20	60
264 g kg <sup>-1</sup> DM	4.74	5.13	56.0	20.0	38.0	60.0	-	0.15	1756	4058
333 g kg <sup>-1</sup> DM	4.20	4.25	62.0	66.2	15.9	16.5	-	-	1550	2240

Table 5. Effect of different dry matter contents (long-term wilting) in alfalfa ensiled in plastic tubes (Schmidt *et al.*, 2001).

	pH			Lactic acid g kg <sup>-1</sup> DM			Acetic acid g kg <sup>-1</sup> DM			NH <sub>3</sub> mg kg <sup>-1</sup> DM		
Day of fermentation:	20	120	270	20	120	270	20	120	270	20	120	270
341 g kg <sup>-1</sup> DM	4.20	4.38	4.51	60.3	79.7	66.8	15.5	17.9	24.0	1498	3455	3394
454 g kg <sup>-1</sup> DM	4.50	4.86	4.90	35.9	55.7	50.6	13.9	09.7	10.3	842	2752	3004
Digestibility (%)	Crude protein			Crude fat			Crude fibre			N-free extract		
341 g kg <sup>-1</sup> DM	71.4			49.8			39.9			76.8		
454 g kg <sup>-1</sup> DM	69.8			48.8			36.8			73.3		

Film thickness (230-280 µm), elongation and strength (3 layers: one UV stable layer, one layer specially for improved strength, and one for low transparency) for the plastic tubes are satisfactory. There is a standard evaluation system for plastic tubes (Csatár *et al.*, 2004), that include the following parameters: thickness, transparency, chroma, slippiness, dart surface, dart deepness, over-ripping factor (transversal and longitudinal), tensile strength (transversal and longitudinal) and elongation till tearing (transversal and longitudinal). In one trial, the average tensile strength range was 24-32 MPa, the rated elongation range was 700- 800%, and the flow limit range was 9.5-10.5 MPa (Csatár *et al.*, 2004).

### 2.3. Conventional ensiling methods: bunker silo, clamp, silage pile

Conventional bunker silos or piles generally are capable of holding 1,000-5,000 Mg in Hungary. High silage density (200-250 DM kg m<sup>-3</sup>) can be achieved within a bunker and a plastic cover helps exclude oxygen and permits proper fermentation. However, these types of silos are expensive to construct (approximately 80,000 EUR/1000 Mg in Hungary), they are not particularly flexible for variable amounts of forage, ensiling is slower than in other systems, and general losses can be relatively high (10-20%) if not managed properly. Cost comparisons of conventional clamp and bag (tubular) systems are shown in Table 6.

Table 6. Distribution of costs required for storage of 5000 Mg alfalfa haylage in a clamp silo and tube silo (Csermely *et al.*, 2002).

Bunker silo		Tubular system	Ag Bag	
			G-6700	G-7000
	% of the total cost		% of the total cost	
Bunker silo	97.3	Filling machine	71.9	78.6
Plastic film	0.55	Plastic film	17.2	12.0
Spreading	0.72	Other cost (transport)	4.6	3.2
Compaction	1.26	Track for filling machine	6.3	6.2
Loader machine	0.15			
			68,220 EUR	91,080 EUR
180,000 EUR 100%			37.9%	50.6%

### 3. Anti-quality factors (undesirable microorganisms and chemicals: mycotoxins) of silages

Apart from contamination of silages with undesired or even pathogenic microorganisms, e.g. *Clostridium tybutyricum*, *Clostridium botulinum*, *Listeria monocytogenes* and *Eschericia coli* (Woolford, 1990), the occurrence of moulds and their secondary toxic metabolites (mycotoxins) require attention as potential factors of poor performance and health disorders in domestic animals. The growth of fungi in silages increases health risk: challenge from spores to the mucosal surface (respiratory diseases) and metabolic disorders associated with ingestion of mycotoxins. Mycotoxins can be carcinogenic, teratogenic, genotoxic, hepato-toxic, nephrotoxic, haemototoxic, immunosuppressive, estrogenic, tremorgenic or mutagenic (Auerbach, 1998). Fungi with the capacity to produce mycotoxins have three critical

environmental requirements during storage: temperature above freezing point, moisture above 200 g kg<sup>-1</sup> and oxygen (Gotlieb, 1997). Rumen protozoa are known to play a significant role in degrading some mycotoxins, but their population might almost totally disappear when animals are fed diets rich in readily fermentable carbohydrates. Therefore, it is critical to prevent or reduce the rate of mycotoxin contamination during harvest and storage.

Many plastic wrapped bales on Irish farms have some mould growth (O'Kiely *et al.*, 1998, 2002) because of higher porosity and 6-8 times more surface area in contact with film compared to clamp silos. Moreover, at times bales are imperfectly sealed resulting in aerobic deterioration. The predominant fungi on baled silage in Ireland are: *Penicillium roqueforti* (42%), *Penicillium paneum* (4%), yeast (13%), *Geotrichum* (3%), *Fusarium* (1%), *Trichoderma* (1%) and *Schizophyllum commune* (20%) (O'Brien *et al.*, 2004 and 2005). In a trial it was confirmed that there was a lower *Schizophyllum* incidence on farms in the north-west region of Ireland, and that low grass DM at ensiling may be a factor that prevents bales being successfully colonized with *Schizophyllum* (O'Kiely *et al.*, 2002). In Scandinavia (Norway), Austria, The Netherlands, Italy, France and northern Germany the predominant mould found in baled silage was also *Penicillium roqueforti* (Auerbach *et al.*, 1998).

Several agricultural practices influence crop contamination in the field (Jouany, 2007) including crop residue, extent and method of tillage, soil fertilizers, planting date, variety selection, use of pesticides or biological control agents, insect- and weed-control, machinery and insect damage to seed, and drought stress. During harvest and post-harvest there is an important role of the physiological stage of plants (earlier harvest results in lower concentration of mycotoxins), the settings on the combine harvester (cutting height, soil contamination), humidity level before and during storage, temperature during storage, pre-cautionary silage making practices (density, porosity, additives, wrapping-sealing technology, transport) and silo wall management (surface area feed out rate) (Jouany, 2007).

#### **4. Management of perennial grasses for bioenergy**

The perennial warm-season grasses, such as switchgrass (*Panicum virgatum*) and big bluestem (*Andropogon gerardii*) are native to the tallgrass prairie and important in forage production, conservation and wildlife habitat (Moser *et al.*, 2004). Another important aspect of switchgrass is its potential use for bioenergy production (Sanderson *et al.*, 2004). Intermediate wheatgrass, a perennial cool-season grass, is important for forage production and for the habitat of livestock throughout the temperate regions. Intermediate wheatgrass is well adapted to areas that receive at least 350 mm of annual precipitation and is productive in marginal land (Asay and Jonsen, 1996). Ross and Krueger (1976) reported that intermediate wheatgrass has superior forage yielding ability, higher than any other grasses in South Dakota USA.

Fertilization and harvest are important management practices for sustainable biomass production of perennial grasses. Harvest management of warm- and cool-season grasses for bioenergy feedstock production should emphasize yield and persistence but not forage quality. A feedstock producer may want to have flexible harvest times for potential fluctuation in future feedstock markets (Sanderson *et al.*, 2004). Also, flexible harvest timing may help a farmer diversify labour. Many researchers have reported optimum harvest timing and frequency for maximum yield and quality of switchgrass biomass feedstock (Sanderson *et al.*, 1999; Vogel *et al.*, 2002; Mulkey *et al.*, 2006). In general, a single harvest during late summer was recommended for maximum yield while a single harvest delayed until late autumn through winter was better for quality of biomass feedstock. Lee and Boe (2005)

suggested a biomass harvest of over-wintered switchgrass. In that system, switchgrass stands could be stockpiled over winter for conservation and wildlife without significant loss of biomass (Owens *et al.*, unpublished data).

Warm-season grasses are N-use efficient and can be adapted to a wide range of soil conditions. However, switchgrass biomass yields generally increase with N fertilizer application. Vogel *et al.* (2002) reported that switchgrass needs approximately 10 to 12 kg N ha<sup>-1</sup> for each Mg of biomass yield in Midwest USA. In South Dakota USA, the optimum N fertilization rate for biomass production and persistence of switchgrass was 56 kg ha<sup>-1</sup> (Mulkey *et al.*, 2006). Big bluestem produces considerable biomass which is comparable to switchgrass and responds to N fertilization rate up to 90 kg ha<sup>-1</sup> (McMurphy *et al.*, 1974; Hall *et al.*, 1983). In general, cool-season grasses respond to N fertilization but this response depends on the availability of water (Smika *et al.*, 1965; Power, 1985). Power (1985) reported that N-use efficiency of intermediate wheatgrass was 51 kg dry matter per kg N in a study conducted in North Dakota USA.

A principal attribute of warm-season grasses such as switchgrass is the potential of high biomass production on marginal lands which are not suitable for conventional row crop production due to high erosion potential (Vogel, 1996). Until now, the major income alternative for producers with marginal or highly-erodible farmland has been to enrol acreage in the federal Conservation Reserve Program (CRP), a costly programme. A shift toward biomass feedstock production with perennial grasses on marginal land would enhance the region's soil organic carbon, overall soil quality, water quality, and wildlife habitat, with the major added economic and rural community benefit of retaining sustainable agricultural systems.

Harvest, storage, and transportation issues must be resolved for these grasses to be successfully developed as bioenergy feedstocks. Since most of these grasses have been utilized in livestock rations, either as conserved forage or as pasture, many producers understand the procedures for harvesting these grasses in large-square or large-round bales. Work is also being done to determine the suitability of these grasses for ensiling. Obviously, any of these species can be ensiled; however, the DM concentration may be 600 g kg<sup>-1</sup> or higher in the field in late autumn (Sanderson *et al.*, 1999; Vogel *et al.*, 2002; Mulkey *et al.*, 2006). Therefore, development of ensiling methods must consider the high DM concentration.

## **5. Developments in biological and chemical silage additives**

### *5.1 Developments in silage fermentation*

Additives are expected to ensure a more efficient fermentation as well as reduce the risk of aerobic deterioration when silages are exposed to air. Moreover, many additives have been developed not just to regulate the ensiling process and prevent aerobic spoilage, but to improve nutritive value of grass silage also.

Organic acids (e.g. formic acid) under difficult ensiling conditions (rainy weather) are the best choice as an aid to preservation and inhibition of harmful microorganisms. Opportunities for different types of additives to improve conservation efficiency may vary with ensiling conditions. Wet crops may be difficult to preserve properly, may ferment extensively and may undergo sizeable losses via effluent. Wilting restricts the extent of fermentation, but may also restrict additive efficacy (O'Kiely *et al.*, 2002). O'Kiely *et al.* (2002) investigated the effect of different additives (formic acid at 3 ml kg<sup>-1</sup> grass; ammonium tetraformate at 4 ml kg<sup>-1</sup> grass;

mixture of formate, sulphite and benzoate at 3 ml kg<sup>-1</sup> grass; and *Lactobacillus plantarum*) in unwilted and wilted (24 hours) grass (*Lolium perenne*) silage (163 g kg<sup>-1</sup> and 259 g kg<sup>-1</sup> DM, respectively). It was shown that the four additives containing formic acid/formate were more effective at improving the preservation and in-silo DM recovery of unwilted grass than the *L. plantarum* additive. The effect of additives on conservation characteristics were generally less with wilted than unwilted forages. *L. plantarum* improved fermentation of wilted grass more than other additives, but silage had reduced aerobic stability at feedout.

Enterobacteria are frequently present on grass at ensiling where contamination with animal manure or soil has occurred. *Escherichia coli* can negatively influence silage fermentation, particularly where the initial rate of pH decline is slow. In a trial by O'Kiely *et al.* (2002) the number of colonies of both inoculated *E. coli* 0157:H7 and indigenous Enterobacteria decreased rapidly to undetectable levels where unwilted precision-chopped grass was ensiled and underwent a rapid lactic acid-dominant fermentation. Altering the ensiling conditions by adding formic acid (850 g kg<sup>-1</sup>, 3 ml kg<sup>-1</sup> grass) increased the rate of decline of both *E. coli* 0157:H7 and Enterobacteria.

Podkowska *et al.* (2005) showed that silages made of endophyte-infected green forage (*Festuca* spp.) treated with a formic acid-based additive (55% formic acid, 24% 4-ammonium formate and 5% propionic acid) had similar quality and nutritive value as untreated endophyte-infected grass silage. However, silage treated with formic-acid had improved aerobic stability.

*Bacterial inoculants* can improve fermentation characteristics by increasing the rate of pH decline and lowering ammonia levels in the silo. Additives containing fibrolytic enzymes provide additional sugar through the breakdown of grass fibre. At present biological additives are preferred because they are non-toxic, non-corrosive to machinery, do not present environmental hazards and are regarded as natural products. Mixtures of different types of additives may improve their overall effect. One of the most popular additives currently available is a bacterial inoculant with cell-wall degrading enzymes. Conditions in the silo may not always be optimal for enzyme function, however. For example, cellulase originating from *Trichoderma reesei* fungi, has an optimal activity between pH 4.8 and 5.2 while that from *Trichoderma viride* is between 4 and 5. Temperature optimum for these cellulase enzymes also differs (Knabe, 1987), and dry matter concentration at ensiling affects enzyme activity. The suggested dry matter concentration for ensiling lucerne is 28-33%, which allows both lactic acid bacteria and cell wall degrading enzymes to work satisfactorily (Schmidt *et al.*, 2001).

## 5.2. Developments in aerobic stability

Aerobic stability of silages may be improved by inhibiting the growth of fungi, moulds and some bacteria after opening the silo. Aerobic spoilage of silage can affect both the efficiency of nutrient utilization and the production of anti-quality compounds such as mycotoxins. Aerobic yeasts can assimilate lactic acid and exacerbate instability upon exposure to air. These yeasts are the cause of heating in spoiled silage and are most active at 20-30°C; therefore it is important to find suitable additives to inhibit fungi and protect the silage upon aerobic exposure. In maize silage, acetic acid bacteria can also play a complementary role in aerobic deterioration. Additives have been used in an attempt to limit aerobic spoilage. Honig *et al.* (1999) suggested that prevention of aerobic instability is primarily contingent on the use of excellent ensiling techniques and the use of appropriate additives.



A number of chemicals have been investigated as silage preservatives such as formic acid, propionic acid and acetic acid. Propionic and acetic acids are fungicidal agents, and high concentrations of propionate and acetate inhibit yeast and mould growth. Undissociated molecules penetrate through passive diffusion into the cells and release  $H^+$  ions, thus lowering the intracellular pH and killing yeast and mould. Filya *et al.* (2005b) found that formic acid-based preservative improved aerobic stability and nutritive value of low dry matter silage by inhibiting yeast and mould growth and reducing pH and  $CO_2$ . Yeasts can also be inhibited effectively by short-chain fatty acids such as acetic acid. The pH-dependent, minimal inhibition concentration of acetic acid for yeasts and moulds is approximately 94 mmol  $l^{-1}$  at 4.0 pH, which is approximately 5.8 g  $kg^{-1}$  DM (Woolford, 1975; Ruser and Klienmans, 2005).

Recent developments have focused on the use of propionic acid bacteria, heterofermentative lactic acid bacteria and a combination of homofermentative lactic acid bacteria with salts such as sorbate, sulphite and benzoate (Davies *et al.*, 2005). Silage additives have had inconsistent effects on aerobic deterioration of grass silages. Filya *et al.* (2005a) demonstrated that *Propionibacterium acidipropionici* improved aerobic stability of silages by inhibition of yeast proliferation. *P. acidipropionici* ( $10^6$  CFU  $g^{-1}$  fresh forage) produced significantly higher levels of propionic and acetic acids, reduced  $CO_2$  level and decreased yeast and mould growth more than *Lactobacillus plantarum* and a combination of *L. plantarum* and *P. acidipropionici*. The combination of *L. plantarum* and *P. acidipropionici* did not effectively protect silage from aerobic deterioration.

Recently, the heterofermentative *L. buchneri* has been shown to improve aerobic stability. *L. buchneri* is a heterolactic bacterium able to ferment lactic acid to acetic acid and 1,2-propandiol under anaerobic conditions. Applied by itself it may negatively affect rate of fermentation, but this can be counteracted by combining it with homofermentative lactic acid bacteria. According to Ruser and Kleiman (2005) *L. buchneri* is primarily effective during the second phase of fermentation when acetic acid and 1,2 propandiol are generated from lactic acid. Ruser and Kleinmans (2005) found that *L. buchneri* improved aerobic stability in grass silage in 27 trials, and was better than with a 5 l  $ton^{-1}$  propionic acid treatment. Acetic acid concentration increased up to 8 g  $kg^{-1}$  DM over untreated silage without a negative effect on feed intake and milk yield. Bach *et al.* (2005) found that silages inoculated with *L. buchneri* had lower maximum temperatures, fungal counts and aflatoxin concentrations after opening compared to untreated silages. There was no significant difference in concentration of deoxynivalenol and zearalenone in inoculated and untreated silages. *L. buchneri* may produce other, as yet, unidentified metabolites with antifungal activity.

A high concentration of lactic acid does not guarantee aerobic stability. Yimin *et al.* (2002) found that *L. casei* and *L. plantarum* improved fermentation quality, but did not inhibit yeast nor aerobic deterioration of Italian ryegrass silages. *L. casei* and *L. plantarum* inoculation inhibited growth of *Clostridia* and aerobic bacteria, but yeast strains isolated from deteriorated silage had a high tolerance to lactic acid and low tolerance to butyric acid. These yeasts were able to grow at low pH and utilize lactic acid for growth, but were inhibited by low concentrations of butyric and propionic acids. Yeasts grew vigorously after opening the silo leading to aerobic deterioration.

White *et al.* (2002) found that silage treated with lactic acid bacteria combined with Na-benzoate and K-sorbate (at 500 and 1000 ppm) produced less  $CO_2$  compared to untreated silage or to silage treated with lactic acid bacteria when exposed to air. The rise in pH was significantly lower in silages treated with a combined additive than in the control and

inoculated silages after 8-days aeration. The combined additive did not adversely affect nutritive value and prevented a decrease in *in-vitro* nutrient digestibility during exposure to air.

Howard *et al.* (2005) investigated different additives containing ammonium tetraformate salt (85%, 3 and 6 ml kg<sup>-1</sup>), *L. plantarum* alone or in combination with sodium benzoate or potassium sorbate (dose: 30 g l<sup>-1</sup>, 5 ml kg<sup>-1</sup>), and salts alone (sodium benzoate and potassium sorbate at 5 ml kg<sup>-1</sup>) in wilted perennial ryegrasses (DM: 372 g kg<sup>-1</sup>). They found that the high rate of the formic acid-containing additive had the greatest positive effect on fermentation and aerobic stability. Including sorbate and benzoate at 5 ml kg<sup>-1</sup> did not improve aerobic stability in their conditions. In another trial, Howard *et al.* (2005) investigated unwilted perennial ryegrasses bred for contrasting sugar contents using the same treatments. They found that the formic acid-based additive had the largest positive effect on fermentation and was the only additive to consistently and significantly improve aerobic stability and reduce aerobic deterioration.

### 5.3 Developments in nutritive value of treated silages and animal performance

Animal performance is the most important aspect in evaluation of a silage additive. During the ensiling process, naturally occurring enzymes (proteases) will cause degradation of true protein (McDonald *et al.*, 1991). Prolonged fermentation in uninoculated silage allows proteolytic bacteria (e.g. Clostridia) to degrade high quality plant protein and convert it to ammonia and nitrates. Therefore, it is essential that the decline in pH is as rapid as possible in the first two or three days of ensilage to stem proteolysis (Webb and Matthews, 1998). In uncontrolled silage fermentation up to 80% of the protein will be broken down (Carpintero *et al.*, 1979). The use of lactic acid-producing bacterial inoculants can significantly reduce the breakdown of protein, leaving higher levels of true protein in the treated forage. Reducing soluble protein levels in the rumen has been reported to improve microbial protein synthesis (Sharp *et al.*, 1994). This overall retention of nitrogen by the animal will lead directly to improved performance, be it live-weight gain in beef animals (Mayne and Steen, 1993) or significant extra milk production in dairy cows, and improves environmental sustainability since less N is excreted in urine and manure.

Steen *et al.* (1995), Keady (1996) and Keady and Murphy (1998) determined the factors affecting DM intake of grass silage by beef cattle. The grass silages were offered *ad libitum* as the sole source of nutrition to growing beef cattle. Key factors affecting silage intake were protein and fibre fractions and the rate and extent of digestion of these components. In a review of silage additives containing sulphuric acid, molasses or enzymes as the main ingredient, Keady (1998) concluded that their use improved silage fermentation but did not significantly improve animal performance. However, in a review of nine studies in which untreated silages were poorly preserved, inoculant treatment increased weight gain in beef cattle and milk yield and quality in dairy cattle.

Grass silage (*Lolium perenne*) inoculated with different lactic acid bacteria (*L. plantarum*, *Pediococcus* spp. and combination) had no beneficial effect on *in-vivo* digestibility, nitrogen retention and blood parameters (b-hydroxy-butyrate, glucose, non esterified fatty acids and urea) in steers. Although all silages underwent a lactic acid dominant primary fermentation, a secondary fermentation proceeded through feedout, with pH-values and concentrations of acetic acid and ammonia-N progressively increasing. The crop contained insufficient fermentable carbohydrates to sustain dominance of lactic acid during primary fermentation. O'Kiely *et al.* (2002) found that inoculation with *L. plantarum*, *Pediococcus* spp. and a combination of the two, improved the fermentation profile of unwilted grass silage, but the



small shift recorded in fermentation products was not sufficient to influence nutritive value in Friesian steers. Lactic acid bacterial inoculation did not affect nitrogen metabolism, blood parameters, rumen degradability of silage DM, NDF or nitrogen, but *Pediococcus* treatments increased the acetate to propionate ratio and tended to increase the non-glucogenic ratio in the rumen fluid. O'Kiely *et al.* (2002) showed that in wet conditions formic acid rather than inoculant (*L. plantarum*) treatment improved silage nutritive value, particularly in the absence of concentrate supplementation. The improved kinetics of rumen digestion and passage in response to formic acid would appear the most likely explanation for the increased silage intake and resultant animal performance.

Zhu *et al.* (2005) found that cell wall degrading enzymes (cellulase) may have a potential for enhancing ruminal degradation of silage, although the benefits were hindered by wilting. *In situ* ruminal degradation of DM and NDF were enhanced by enzyme (cellulase) addition at initial incubation time, while the effect on DM degradation diminished in wilted Italian ryegrass.

Feeding of aerobically unstable silage negatively affects rumen fermentation in dairy cows, causing alimentary disorders associated with low feed intake, poor rumination and nutrient utilization. Aerobically deteriorated silage increased ammonia concentration and rumen pH but reduced acetic acid concentration and had a negative effect on microfauna in the rumen (Dvoracek and Dolezal, 2003). Filya *et al.* (2005b) found that a formic acid-based preservative improved not only aerobic stability but nutritive value of low dry matter silage.

Based on this review it is apparent that silage additives may improve the feeding and nutritive value of grass silages but their usage cannot replace professional silage management.

## **6. Required further research and development**

### *6.1 Silage*

Anti-quality aspects require further investigation and development especially in baling systems (higher density, lower porosity, more efficient wrapping with lower gas permeability, prevention of plastic film damage) to reduce animal health risk.

As a result of increasing energy costs and environmental concerns, there is a need to investigate the energy efficiency of equipment with high output capacity while reducing harvest and storage losses (Forristal and O'Kiely, 2005).

Success of ensiling strongly depends on forage characteristics, especially dry matter content at ensiling. There is no time during harvest to wait for lab analyses, so improvements in accuracy of in-field analysis techniques for estimation of dry matter content and other forage parameters would be extremely helpful. Near infrared (NIR) techniques may help by allowing for real-time on-harvester fresh weight yield sensing, dry matter determination, forage quality characterization, and prediction of feed intake based on silage characteristics of material going into the silo. This is also important in development of biomass feedstocks if ensiling is the desired harvest method. There is also a need for rapid (in-field) and inexpensive mycotoxin analysing methods.

### *6.2 Biomass*

With the high price of oil, it is imperative that research on perennial grasses for biomass continues. Identification of appropriate species for given locations and environments is

critical to optimize yields and environmental sustainability. Furthermore, matching the species to the landscape will allow producers to provide a diverse set of biomass species for biomass energy production.

Scaling-up to production-size fields will be necessary to evaluate potential biomass species at the farm level. Economics of these practices must also be considered in order to provide producers with incentives to grow perennial grasses for energy.

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## **Session 3A**

# **Organic production and nutrient flows**



# Towards nitrogen self-sufficiency in mixed crop organic dairy systems: legumes and protein-rich plant contributions

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## Abstract

Nitrogen is known to be the limiting factor of productivity in organic systems. In low-input organic, mixed-crop dairy systems (MCDS) at the Mirecourt INRA station, studies are conducted on the contribution of associated cereal/protein-rich plant crops and concentrate mixes to the nitrogen self-sufficiency of the system. An agronomic and zootechnical analytical trial was established with nine types of cereal and protein-rich plants, and associated cereal/protein rich plant crops were compared in plot trials in 2006 and 2007. The valorization of three cereal/protein concentrate mixes by 24 lactating dairy cows was tested in a zootechnical experiment during winter 2007. Dairy cows were each fed 4 kg d<sup>-1</sup> of the mix, 8 kg DM d<sup>-1</sup> of lucerne/ cocksfoot hay, and permanent grassland hay *ad libitum*. Associated cereal/protein rich plant crops produced a greater and more stable yield (3.0 t ha<sup>-1</sup>) than pure cereals (2.2 t ha<sup>-1</sup>) but the proportions of protein-rich grains in the mixes were highly variable (26 to 62%). Dairy cows' feed requirements were satisfied by the three diets. Cereal/protein concentrate mixes had a low impact on dairy cows' performance because of the high contribution of forages to the diet.

Keywords: mixed-crop dairy systems, low input organic systems, nitrogen self-sufficiency, legumes, protein-rich plants

## Introduction

In the Mirecourt INRA experimental farm, situated in the Vosges lowland, the agro-ecological sustainability of two low-input complementary organic dairy systems is being evaluated: a grassland system (GS) and a mixed-crop dairy system (MCDS). Agro-ecological evaluation of these systems deals with their productivity, and their impacts on five environmental components: water and air quality, soil quality, biodiversity and energy consumption. In these systems, nitrogen is known to be one of the main limiting factors of productivity. The situation might be worse in MCDS, compared with GS, because of the presence of annual crops in the rotations exporting many minerals and low restitutions. The MCDS is composed of 50 ha of permanent grassland and 110 ha of rotation crops. The dairy herd comprises 55 to 60 dairy cows, and heifers for replacement. Two crop rotations are tested on the farm: 8- and 6-year successions maximizing cereal crops in order to be self-sufficient in straw. The first phase of rotation consists of mixed swards composed of lucerne/ cocksfoot, and clovers/ ryegrass/ fescue, for the 8- and 6-year crop rotations respectively. To compensate for the lack of restitutions by manure, averaging 800 t y<sup>-1</sup> for the 160 ha, the 5-years of annual cereals following the mixed swards, needs another source of nitrogen in the 8-year crop rotation. Therefore, one year of associated cereals/protein-rich (legume) crop is introduced. The dairy herd, calving in late summer and autumn, has a high level of production and feed requirement during the indoor feeding period. The challenge is to assure the sustainability of the dairy herd by covering their needs using forages and concentrates grown in the system. We will focus on the contribution of associated cereal/protein-rich plant crops and concentrate mixes to the nitrogen self-sufficiency of the

MCDS, considering (i) the nitrogen they contribute to the soil that might be available for the next crop, and (ii) their contribution to meeting the protein requirements of the dairy cows.

## Material and methods

Two analytical trials have been set up to consider nitrogen self-sufficiency in the MCDS. Agronomic and environmental performances of associated cereals and protein-rich (legume) crops were evaluated in small plot trials, inserted into the 8-year crop rotation of the MCDS, in 2005/2006 and 2006/2007, following the same protocol. Nine types of winter cereals, protein rich plant and associated cereal/protein rich plants were compared in homogenous pedoclimatic conditions each year (2005/2006: clay loam soils on dolomite; in 2006/2007: silty clay soils on dolomite). These were: wheat, triticale, pea, wheat/pea, triticale/pea, oat/pea, triticale/vetch, oat/vetch and triticale/oat/pea/vetch. Measurements were made of: sowing density, seedling counts before and after winter period, evidence of stress and disease, weeds, yield at harvesting, and nitrogen mineral concentration in the 0-90 cm soil several times each season (sampled in four horizons: 0-15, 15-30, 30-60 and 60-90 cm). Experimental results were analysed by descriptive statistics.

Zootechnical performances of lactating dairy cows from the MCDS fed by 3 different cereals/protein concentrate mixes were compared during the winter of 2007. Twenty-four mid-lactation dairy cows (milk production:  $20.7 \pm 3.6$  kg cow<sup>-1</sup> d<sup>-1</sup>; body weight :  $648.4 \pm 52.8$  kg cow<sup>-1</sup>; body condition score:  $1.6 \pm 0.4$ ) were fed with 8 kg DM cow<sup>-1</sup> d<sup>-1</sup> of lucerne/cocksfoot hay, permanent grassland hay *ad libitum*, and 4 kg cow<sup>-1</sup> day<sup>-1</sup> of one of the three different cereal/protein concentrate mixes (oat/horse bean, barley/lupin, triticale/pea). The experiment was organized in a Latin square design. Three groups of 8 cows were set up by taking into account the breed (50% of Holstein and 50% of Montbeliard) and the parity (50% of primiparous and 50% of multiparous). The trial was composed of 3 experimental periods of 4 weeks: 2 weeks of adaptation to the diet and 2 weeks of measurements. All measurements were carried out at the individual cow level: daily intake, daily milk production, milk fat, milk protein and urea content; body weight and body condition score at the end of each experimental period. Experimental results were analysed by analysis of variance.

## Results and discussion

In 2006 and 2007, yields of associated cereal/protein rich plant crops (2006:  $3.5$  t ha<sup>-1</sup>; 2007:  $2.6$  t ha<sup>-1</sup>) were higher than yields of cereals (2006:  $2.5$  t ha<sup>-1</sup>; 2007:  $1.9$  t ha<sup>-1</sup>) and were more stable than yields of single-species crops. The proportion of protein-rich grain varied from 42 to 62% of the total weight of the mixes in 2006, and from 26 to 62% in 2007. Variations of yields and proportions of cereals and protein-rich grain in the mixes between 2006 and 2007 are partly explained by the loss of germinating seeds between sowing and the end of winter in 2007 due to a high rainfall in October 2006 (200% higher than the 15-year average).

Nitrogen fixation and restitution to the soil after harvesting (July 2006 and 2007) was higher with associated cereal/protein rich plant crops ( $35$  kg N ha<sup>-1</sup>) than cereals crops ( $21$  kg N ha<sup>-1</sup>) in 2007, but equal in 2006 ( $\approx 45$  kg N ha<sup>-1</sup>). Nitrogen fixation by legumes depends on the nitrogen concentration in the soil, which was higher at the end of winter 2006 ( $56$  kg N ha<sup>-1</sup>) than at the end of winter 2007 ( $\approx 20$  kg N ha<sup>-1</sup>), due to manure spreading in autumn 2005 and to high leaching due to the high rainfalls of October 2006. In 2007, nitrogen restitution to the soil was higher with pure protein-rich crops ( $57$  kg N ha<sup>-1</sup>) than with associated cereals/protein-rich plant crops ( $35$  kg N ha<sup>-1</sup>). This might be explained by the use of fixed nitrogen by the associated cereal and by a lower proportion of legumes in the associated crops.

Dairy cows fed by the three different diets had the same milk production and milk fat content (Table 1). Milk protein content was  $0.5 \text{ g kg}^{-1}$  ( $P < 0.001$ ) lower for cows fed by oat/horse bean concentrate mix compared with cows fed the other two diets. Energy and PDI balances were positive, meaning that the feed requirements of the dairy cows were totally satisfied by the three diets. Diets had a PDI/UFL balance significantly ( $P < 0.0001$ ) different, showing an increasing proportion of digestible protein in diets from diet containing barley/lupin to diet containing triticale/pea and to diet containing oat/horse bean. This higher proportion of protein and lower level of starch ( $-450 \text{ g cow}^{-1} \text{ day}^{-1}$ ) in the diet containing oat/horse bean concentrate mix may explain the lower level of milk protein content for cows fed that diet compared with cows fed by the other diets. The differences of protein content of the diets are explained by the heterogeneous composition of mixes harvested and used as concentrates on the MCDS. The proportions of protein grains in the mixes were 2.3% in barley/lupin mix, 46.9 % in oat/horse bean mix and 59.9% in triticale/pea mix.

Concentrate mixes have a low impact on dairy cow performances of the MCDS: forages, including a rich hay like lucerne/cocksfoot hay, supply on average 77% and 89% of energy and protein requirements of dairy cows, respectively.

Table 1. Effects of the 3 diets (with  $8 \text{ kg DM cow}^{-1} \text{ day}^{-1}$  lucerne hay, permanent grassland hays *ad libitum* and  $4 \text{ kg cow}^{-1} \text{ day}^{-1}$  of the three different cereal/protein concentrate mixes (oat/horse bean, barley/lupin, triticale/pea)) on dairy cow performances, balance between dairy cows intake and their needs and diet balance.

Variables	Diets			Significance ( <i>P</i> =)
	Oat/Horse bean	Barley/Lupin	Triticale/Pea	
<i>Dairy cows performances</i>				
Milk (kg)	20.4	20.1	20.3	0.3921
Fat (g kg <sup>-1</sup> )	41.0	41.4	41.6	0.1091
Protein (g kg <sup>-1</sup> )	31.4 <sup>A</sup>	32.0 <sup>B</sup>	32.2 <sup>B</sup>	0.0010
<i>Balances</i>				
Energy (UFL <sup>E</sup> d <sup>-1</sup> )	0.89	0.96	0.95	0.8801
PDI <sup>F</sup> (g d <sup>-1</sup> )	307 <sup>Aa</sup>	228 <sup>B</sup>	270 <sup>Ab</sup>	0.0027
<i>Diet balance</i>				
PDI/UFL <sup>G</sup> (g UFL <sup>-1</sup> )	105.97 <sup>A</sup>	101.22 <sup>B</sup>	104.01 <sup>C</sup>	0.0001

A, B, C:  $P < 0.01$  a, b:  $P < 0.05$

E: Difference between the Net energy in feed unit for milk ( $1 \text{ UFL} = 7.115 \text{ MJ NE}$ , INRA, 1989) intake and the net energy requirements.

F: Difference between the Digestible Proteins in the Intestine (PDI) intake and the PDI requirements (INRA, 1989).

G: Balance of the energy and protein daily diet: the diet is well balance for dairy cows when it is around  $100 \text{ g PDI/UFL}$ .

## Conclusion

In a low-input organic mixed crop dairy system, an associated cereal/protein-rich plant crop assures a stable yield of grain (and straw) and provides nitrogen to the soil, available for the next crop. But the composition of the harvested mix is unpredictable. In our trial concentrate mixes have a low impact on the performance of dairy cows because of the low level of mixes in the diet and the presence of rich hay containing legumes. These concentrate mixes might have a better contribution when they complement poor forages like permanent grassland hays.

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# Effects of different stocking rates with dairy cows on herbage quality in organic farming

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## Abstract

In order to identify the effects of stocking rate of grazing dairy cows on herbage quality in organic farming, grazing experiments were conducted from 2004 to 2006. All lactating Holstein cows of one herd were divided into two groups during vegetation periods. The paddocks of the rotational pasture were split in a way that the low stocking rate group (LSR) had 15% more pasture area than the high stocking rate group (HSR). Post-grazing sward height for HSR was decisive for the simultaneous change of the sub-paddocks. Annual stocking rates for HSR were between 2.0 and 2.3 cows ha<sup>-1</sup>. At regular intervals two grass strips per stocking rate were cut and sampled in the paddock to be grazed next. No differences were revealed concerning the strongly varying pre-grazing herbage mass with 1,235 (HSR) and 1,165 kg DM ha<sup>-1</sup> (LSR). In the offered herbage mass for the LSR group, significantly lower ash, crude protein, absorbable protein in the duodenum based on energy or nitrogen available in the rumen, net energy for lactation, as well as higher neutral detergent fibre and acid detergent fibre contents were detected. Sugar values were unaffected. Referring to herbage mineral concentrations, only the potassium concentration was significantly lower in herbage for the LSR group. The allocation of a larger grazing area without pasture topping leads to a lower pasture quality, mainly from mid June to the end of August.

Keywords: pasture, herbage quality, stocking rate, organic farming, dairy cows

## Introduction

In Switzerland, pasture-based milk production systems offer an optimal possibility to take advantage of the climate providing ample and regular grass growth, as well as minimizing the impact of topographic disadvantages. It is also well established that grazed grass is a low cost forage with a high nutrient value. According to the directives of Swiss organic farming, ruminants need to have access to pasture during the vegetation period to ensure a natural feeding system as well as for animal welfare. The efficiency of pasture use under organic farming, which in most cases implies restricted nitrogen supply, can still be improved.

The objective of this research was to study the effects of two different stocking rates with dairy cows on herbage quality in organic farming.

## Materials and methods

The investigations were conducted from 2004 to 2006 on the farm 'l'Abbaye' in Sorens, Switzerland (46°39.767' N; 7°3.143' E). The conversion of this farm to organic farming started in 2003 and was officially finished in 2005. During the three vegetation periods (April to November) means of temperature (site at Payerne, 490 m above sea level (a.s.l.), and *ca.* 2 °C higher than around the farm) and sums of precipitation were: 13.2 °C, 819 mm; 13.3 °C, 748 mm, and 14.2 °C, 991 mm (MeteoSwiss). All lactating Holstein cows (on average 60 cows) were mainly fed pasture, and during the three vegetation periods were divided into two groups: high stocking rate (HSR) and low stocking rate (LSR). The sixteen paddocks of the rotational pasture system (total pasture area 33 ha, 800 to 900 m a.s.l.) were split such that

LSR had 15 per cent more pasture area available than HSR. The post-grazing sward height (POGSH) for HSR, measured with a rising plate meter (Filip's folding plate pasture meter, Jenquip, NZ) was decisive for the simultaneous change of the sub-paddocks for both groups. Every two weeks in 2004, and once per week in 2005 and 2006, respectively, two grass strips per stocking rate were cut (average cutting level 8.7 Units, 1 unit corresponds to a compressed sward height of 0.5 cm) and sampled in the paddock to be grazed next to evaluate the pre-grazing herbage mass (PRGHM) and herbage quality. Ash, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), total sugar, calcium (Ca), phosphorus (P), magnesium (Mg) and potassium (K) were analysed. Net energy for lactation (NEL) and absorbable protein in the duodenum, based on energy (APDE) or nitrogen (APDN) available in the rumen, were calculated according to RAP (1999). The signs test for paired samples was applied to compare data.

## Results

Table 1. Pre-grazing herbage mass, botanical composition and mineral content.

	HSR			LSR			<i>P</i>
	Median	Minimum	Maximum	Median	Minimum	Maximum	
PRGHM (kg DM ha <sup>-1</sup> )	1235	317	2774	1165	382	3234	-
Grasses (per cent)	71	42	95	72	41	96	-
Legumes (per cent)	12	2	55	15	1	55	-
Herbs (per cent)	5	1	45	6	0	50	-
Calcium (g kg <sup>-1</sup> DM)	5.3	4.4	10.3	6.6	3.6	9.6	-
Phosphorus (g kg <sup>-1</sup> DM)	4.4	3.7	5.6	4.4	3.4	5.5	-
Magnesium (g kg <sup>-1</sup> DM)	2.0	1.5	2.6	1.9	1.5	2.6	-
Potassium (g kg <sup>-1</sup> DM)	34.4	27.6	41.8	32.8	21.8	38.3	*

\*Significant at  $P < 0.05$ .

Table 2. Herbage quality 2004-2006.

Period	1			2			3			Overall minimum (min.) and maximum (max.)			
	Turnout-mid June			Until end of August			Until turn-in			HSR		LSR	
	$x_{\text{HSR}}^a$	$x_{\text{LSR}}^a$	<i>P</i>	$x_{\text{HSR}}^a$	$x_{\text{LSR}}^a$	<i>P</i>	$x_{\text{HSR}}^a$	$x_{\text{LSR}}^a$	<i>P</i>	min.	max.	min.	max.
Ash <sup>b</sup>	104	102	-	103	99	*	120	118	-	81	197	84	167
NDF <sup>b</sup>	439	434	-	472	487	*	463	461	-	352	534	325	592
ADF <sup>b</sup>	255	241	-	284	304	*	273	278	-	206	329	214	360
CP <sup>b</sup>	173	164	-	167	143	**	185	180	-	132	229	113	224
Sugar <sup>b</sup>	93	97	-	64	61	-	51	55	-	42	146	41	134
APDE <sup>b</sup>	104	102	-	100	96	*	103	104	-	91	120	86	116
APDN <sup>b</sup>	115	109	-	110	94	**	123	120	-	87	152	74	149
NEL <sup>c</sup>	6.2	6.2	-	5.9	5.7	*	5.9	5.9	-	5.4	7.1	5.0	6.8

\*Significant at  $P < 0.05$ , \*\* significant at  $P < 0.01$ .

<sup>a</sup> $x_{\text{HSR}}$  median for HSR,  $x_{\text{LSR}}$  median for LSR.

<sup>b</sup>g kg<sup>-1</sup> DM.

<sup>c</sup>MJ kg<sup>-1</sup> DM.

From 2004 to 2006 the average stocking rates during the vegetation periods were for HSR 2.0, 2.3 and 2.3, and for LSR 1.7, 2.0 and 1.9 cows ha<sup>-1</sup>. The average POGSH for HSR, as a criterion for simultaneous change of paddocks, and as an indicator for grazing severity for HSR, were 10.7 (2004), 9.7 (2005) and 9.0 Units (2006). The strongly varying PRGHM, botanical composition and mineral concentrations are shown in Table 1. No differences were revealed concerning the PRGHM. Proportions of grasses, legumes and herbs in the sward were similar, and remained similar, in both treatments. Herbage quality analyses of the herbage on offer showed that of Ca, P, Mg and K concentrations, only the K concentration was significantly lower in LSR. From mid June to end of August significantly lower values

for ash, CP, APDE, APDN and NEL, as well as higher NDF and ADF values, were detected in the offered herbage for LSR. Sugar values were unaffected by the different stocking rates.

## Discussion

As concentrate feeding is limited in organic farming, the quality of the forage is extremely important for covering the requirements of dairy cows for milk production. The average nutritive value of herbage for HSR was relatively high, with a crude protein concentration of 174 g CP kg<sup>-1</sup> DM, and 459 g NDF kg<sup>-1</sup> DM, and 6.0 MJ NEL kg<sup>-1</sup> DM, but considerable seasonal variation appeared. The CP and NDF values were comparable to the values reported in Kuusela *et al.* (2002) and Kuusela (2004), although conditions are quite different. Over the three experimental years the average Ca, P, Mg and K concentrations were similar to the data from Kuusela (2006) and are not, overall, sufficient to meet the requirements of dairy cows. Lower values for CP, APDE APDN and NEL, as well as higher NDF and ADF values for offered herbage in the LSR group, show a decreasing herbage quality during the second period from mid June to the end of August, with too lax grazing without pasture topping (cutting of pasture residues) and stocking rates that are too low.

## Conclusions

The average nutritive value of herbage for HSR was relatively high, but considerable seasonal variations appeared. Higher pasture area allowance without pasture topping leads to a lower herbage quality from mid June to the end of August.

## Acknowledgments

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# Productivity and N-leaching in organic dairy grass-arable crop rotations

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## Abstract

The increasing size of Danish organic dairy farms means that uniform grazing of all cropped land becomes inexpedient due to long distance to the milking facilities. We investigated nutrient dynamics and feed production in two dairy crop rotations with internal differences in proportions of grazing and cutting land. One six-year crop rotation represents land close to the farm buildings (barley (*Hordeum vulgare* L.) undersown with grass-clover (*Lolium perenne* L.–*Trifolium repens* L.), 4 years of grass-clover, spring barley/catch crop) and another represents land further away (barley undersown with grass-clover, 2 years of grass-clover, barley/catch crop, maize (*Zea mays* L.)/catch crop, lupin (*Lupinus angustifolius*)/catch crop). In each of the crop rotations there were five management treatments for grazing/cutting strategy and manure application. Results from the first experimental year show that herbage production was high in year 1-4 of grass-clover. Nitrate leaching in the crop rotations were highest in grazed and manured 2-4-year-old grasslands, but also following maize and lupin considerable losses occurred despite the presence of catch crops. Following grassland cultivation, a barley silage crop undersown with Italian ryegrass (*Lolium multiflorum* L.) reduced leaching to a minimum.

Keywords: organic farming, nitrate leaching, manure, grazing, cutting, grass-clover productivity

## Introduction

Historically, a large part of organic milk has been produced on smaller farms with maximum integration of animal husbandry and plant production through grazing of the entire crop rotation. An increased proportion of grass-clover in combination with an ongoing structural development in the size of dairy farms, conventional as well as organic, has lead to high proportions of grassland near the farms as uniform grazing of all cropped land becomes inexpedient due to long distance from the milking facilities. On Danish organic dairy farms we already experience grass-intensive crop rotations located close to farm buildings and with grass-clover pastures of longer duration than the 2-3 years that have previously been common. This development has implications. A concentration of grazed grassland near the farm creates loss of fertility furthest from the farm and accumulation of nutrients near the farm, to an extent that may increase losses, e.g. of nitrogen, if not efficiently utilized. This is especially important in areas with sandy soils and high winter rainfall where a large proportion of organic dairy farms are located in Denmark. However, the longer duration of grasslands may also provide an opportunity to control nutrient losses due to less frequent grassland cultivation. The theme of project reported in this paper is grass-clover leys as an integrated part of organic dairy farms. The focus is on management strategies with the purpose of overcoming the above-stipulated shortcomings by manipulating grassland frequency and grazing intensity (and nutrient load). Results from the first experimental year are presented.

## Materials and methods

Two crop rotations were established on a loamy sand soil in an existing grass-arable system at the Research Centre Foulum. One rotation represents land close to the farm buildings



(barley/grass-clover, 4 years of grass-clover and barley/catch crop) and another represents land further away (barley/grass-clover, 2 years of grass-clover, barley/catch crop, maize/catch crop and lupin/catch crop). The area of each crop was *ca.* 0.3 ha. In all grass-clover leys five grassland treatments were made varying in nutrient load and grassland management (Table 1). The other crops in the crop rotations were used as indicators of residual effects. Each combination of crop rotation, crop and grassland treatment is present in duplicate each year. Adjacent to the crop rotations, permanent grassland established in 1993 was used as a reference for crop production with the five grassland management treatments established. In the crop rotations (not the permanent grassland) leaching of nitrogen was estimated from nitrate concentrations in soil water sampled by means of ceramic suction cups, and the accumulated nitrate leaching was calculated after modelling the water balance. Leaching nitrate was analysed using a general linear mixed model on log-transformed data, but results are presented as arithmetic means.

Table 1. Grassland management treatments. Grazing plots grazed continuously by heifers.

Management treatments
1 Grazing regime with cattle manure application in spring (100 kg total-N ha <sup>-1</sup> )
2 Grazing regime without manure application
3 One cut followed by a grazing. Cattle manure application in spring (100 kg total-N ha <sup>-1</sup> )
4 Cutting with cattle manure application (200 kg total-N ha <sup>-1</sup> , ½ in spring and ½ after 1st cut)
5 Cutting regime without manure application

## Results and discussion

Annual grass-clover production in 2006 is shown for cut grassland in Table 2. Within the period years 1-4, there was little variation in dry matter (DM) yield and the yield increase caused by manure application was only 5-15%. The explanation for this relatively modest effect of manure was a compensating increase in clover content in unmanured grassland (results not shown). As an effect of the high clover content, especially in the second and third cuts, the N yields from manured and unmanured grasslands were almost identical. The effect of manure was greater in the 13-yr-old grassland probably due to a lower proportion of clover.

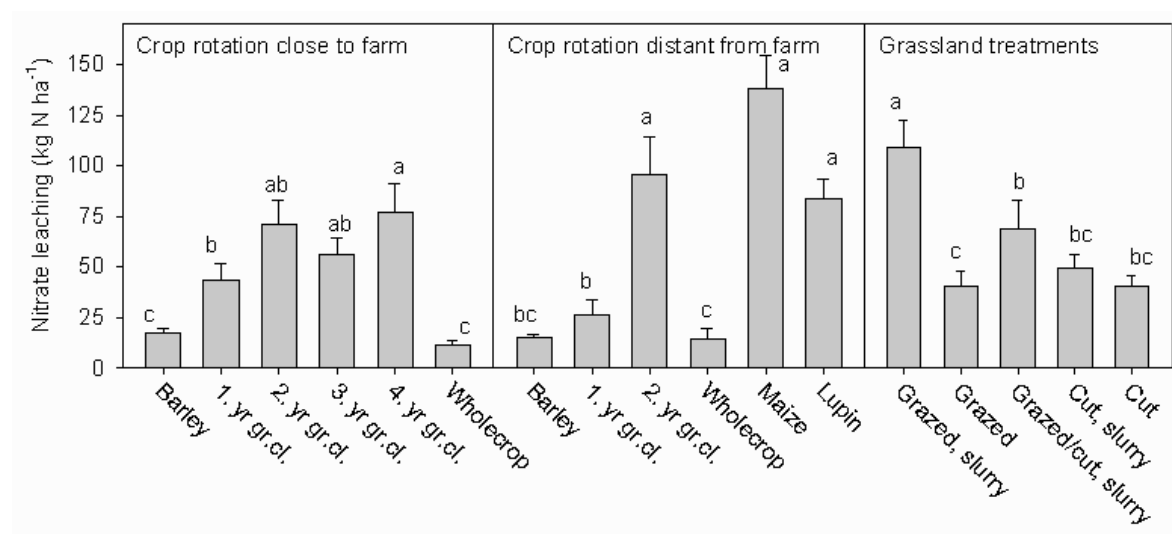
Table 2. Annual herbage and nitrogen yield for 2006 in cutting regimes ( $\pm$  SE).

Grass-clover prod. year	Herbage yield (Mg DM ha <sup>-1</sup> )			Nitrogen yield (kg N ha <sup>-1</sup> )		
	0 N	200 N	Increase	0 N	200 N	Increase
1st	9.4 $\pm$ 0.4	10.8 $\pm$ 0.4	15%	268 $\pm$ 7	263 $\pm$ 8	-2%
2nd	10.0 $\pm$ 0.4	10.8 $\pm$ 0.2	7%	277 $\pm$ 13	285 $\pm$ 11	3%
3rd	9.7 $\pm$ 0.2	10.1 $\pm$ 0.3	5%	269 $\pm$ 10	273 $\pm$ 8	2%
4th	9.5 $\pm$ 0.1	10.6 $\pm$ 0.2	11%	260 $\pm$ 4	279 $\pm$ 18	7%
13th	7.1 $\pm$ 1.1	8.4 $\pm$ 1.2	18%	175 $\pm$ 23	199 $\pm$ 25	14%

Nitrate leaching (Figure 1) in the crop rotation close to the farm was mainly in the grasslands, and generally nitrate leaching was lowest in the first production year. In both crop rotations the barley wholecrop, undersown with Italian ryegrass, was very efficient in accumulating N following spring-ploughed grassland. Therefore, leaching losses at this place in the crop rotations were at a very low level, as earlier demonstrated by Hansen *et al.* (2007). Distant from the farm leaching losses following maize and lupin were considerable, despite both crops having been followed by a catch crop. Maize was undersown with a ryegrass/winter rape mixture and lupin was followed by winter rye. Nitrate losses in grasslands depended on both grazing and manure treatment. Nitrate concentration peaks of random nature were experienced in grazed plots probably caused by urination of grazing cattle. Figure 1 shows the average of all grasslands but the same pattern appeared more or less in all grasslands. Highest

nitrate leaching was found following the grazing regime with manure application, but a considerable drop was observed when avoiding the manure application. A drop was also observed when removing a first cut before start of grazing, although this was not as efficient as avoiding manure. In cut grassland manure application did not influence nitrate leaching. Previously it has been found that leaching losses were increased in fertilized and grazed grassland at this location (Eriksen *et al.*, 2004).

Figure 1. Annual mean nitrate leaching winter 2006-2007. Left and centre: individual crops in the two crop rotations, average of grassland regimes. Right: grassland regimes, average of grassland age and crop rotation. Error bars:  $\pm$ SE. Bars with the same number within each plot are not significantly different ( $P < 0.05$ ).



## Conclusions

Results from the first experimental year show that herbage production was high in years 1-4 of grass-clover and with an N fertilizer response corresponding to 2-7 kg DM kg<sup>-1</sup> N. Nitrate leaching in the crop rotations was highest in grazed and manured grasslands 2-4 years old, but also following maize and lupin considerable losses occurred despite the presence of catch crops. Following grassland cultivation, a barley whole crop for silage undersown with Italian ryegrass reduced leaching to a minimum.

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# In-field N transfer, build-up, and leaching in ryegrass-clover mixtures

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## Abstract

Two field experiments investigating dynamics in grass-clover mixtures were conducted, using <sup>15</sup>N- and <sup>14</sup>C-labelling to trace carbon (C) and nitrogen (N) from grass (*Lolium perenne* L.) and clover (*Trifolium repens* L. and *Trifolium pratense* L.). The leaching of dissolved inorganic nitrogen (DIN), as measured in pore water sampled by suction cups, increased during the autumn and winter, whereas the leaching of dissolved organic nitrogen (DON) was fairly constant during this period. Leaching of <sup>15</sup>N from the sward indicated that ryegrass was the direct source of less than 1-2% of the total N leaching measured, whereas N dynamics pointed to clover as an important contributor to N leaching. Sampling of roots indicates that the dynamics in smaller roots were responsible for N and C build-up in the sward, and that N became available for transfer among species and leaching from the root zone. The bi-directional transfer of N between ryegrass and clover could, however, not be explained only by root turnover. Other processes like direct uptake of organic N compounds, may have contributed.

Keywords: DON leaching, N transfer, field experiment, isotope labelling

## Introduction

Efficient utilization of nutrients in agricultural systems is essential in order to meet a sustainable production of food and reduce environmentally harmful losses. Grass-clover mixtures are a key crop on e.g., dairy farms, supplying high quality fodder and at the same time building up soil fertility. The main input of N in such systems is derived from N<sub>2</sub>-fixation of clover, which means that understanding the below ground dynamics is essential to reduce N loss. In recent years there has been a growing awareness that investigations of N losses have to include organically bound N (DON) as this can contribute significantly to the total N loss.

The aim of the present studies was to investigate the N dynamics in perennial ryegrass-clover mixtures that lead to transfer among species, C and N build-up, and DOC, DIN, and DON leaching.

## Materials and methods

In the two field experiments PVC cylinders (ø 30 cm) were inserted into existing grass-clover swards to confine the system under investigation. Beneath the cylinders Teflon suction cups were installed in order to collect pore water percolating the cylinders. N dynamics was studied by <sup>15</sup>N and <sup>14</sup>C labelling, added in one experiment directly in the field through leaf-labelling (Rasmussen *et al.*, 2007), and in the other experiment via <sup>15</sup>N- and <sup>14</sup>C-labelled residues, which were incubated in the cylinders (Rasmussen *et al.*, submitted). The leaf-labelling experiment lasted for three months during the summer, whereas the residue incubation experiments lasted for 16 months, covering two growth seasons and one autumn-winter period with high pore water leaching. In both experiments N concentration and <sup>15</sup>N- and <sup>14</sup>C-enrichment in leaf material, roots, soil and percolating pore water was determined. The

latter measured in form of both DIN and total dissolved N (TDN), which allow for the calculation of DON by simple subtraction.

## Results and discussion

The two experiments gave many interesting findings of which two concerning the  $^{15}\text{N}$ -data will be discussed here. Of major agronomic importance is the transfer of N between the two species. The experiment with direct leaf-labelling of either ryegrass or clover confirmed that N transfers in such mixtures are bi-directional (Høgh-Jensen and Schjoerring, 2000) with a net-transfer from clover to ryegrass. We found that 40% of the N in ryegrass originated from clover, whereas 5% of N in clover came from the companion ryegrass. The mode of N transfer has been discussed in the literature (e.g. Goodman, 1991; Lipson and Nasholm, 2001; Moyer-Henry *et al.*, 2006).

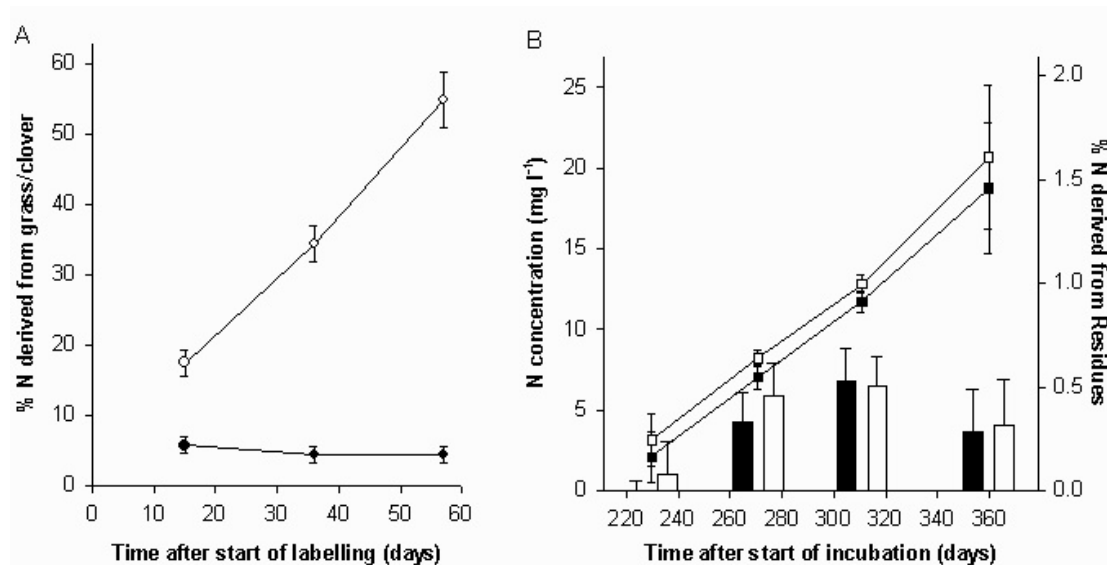


Figure 1. A: Transfer of N from grass to clover (●) and from clover to grass (○) with time in the leaf-labelling experiment. Error bars shows standard error (s.e.) for nine replicates. B: Curves show leaching of DIN (■) and TDN (□), and bars show  $^{15}\text{N}$ -enrichment as % N derived from Residues of DIN (black bar) and TDN (white bar) in the residue incubation experiment. The plot show as an example the data for the incubation of  $^{15}\text{N}$ -labelled clover roots. Error bars shows standard error (s.e.) for four replicates.

Our results (Figure 1A) indicate that different modes of N transfer are in action when comparing the bi-directional transfer. Transfer of  $^{15}\text{N}$  from ryegrass to companion clover was highest initially after leaf-labelling and thereafter decreasing, which points to fast processes like clover uptake of inorganic or organic root depositions from grass or direct root-root interconnections via mycorrhizae. Transfer of  $^{15}\text{N}$  from clover to ryegrass was fairly high immediately after leaf-labelling, meaning that the same process above was in play, but as the  $^{15}\text{N}$  transfer increased markedly with time, it is likely that clover root turnover is of major importance for the total N transfer.

The environmental concern regarding DIN and DON leaching was studied in the experiment with dual-labelled residues (white clover root, ryegrass root, white clover leaves, ryegrass leaves). The leaching of DON and DIN did not seem to come directly from the labelled residues. Instead we found a positive correlation between the uptake of  $^{15}\text{N}$  by ryegrass in the growing season prior to the period of leaching in autumn and winter, and the  $^{15}\text{N}$ -enrichment

of DON and DIN. The results point to turnover of ryegrass roots to be a main source of the  $^{15}\text{N}$ -enriched DON and DIN percolating from the cylinders; this not exceeding 1.5% though. The total N loss during autumn and winter in the studied system was  $10 \text{ kg N ha}^{-1}$ , of which 15% was found as DON. During autumn and winter there was an increasing leaching of DIN, whereas the leaching of DON was fairly constant (Figure 1B). Interestingly, there was also a shift in the  $^{15}\text{N}$ -enrichment of N lost from DON in autumn to DIN by the end of winter.

## Conclusions

The fairly slow process of clover root turnover was found to be the most important regarding N transfer to companion grass, although faster processes were also in play. Transfer in the direction from grass to clover was fast and therefore expected to be in the form of either clover uptake of root deposits or direct root-root interconnections.

Turnover of ryegrass roots was directly related to the leaching of  $^{15}\text{N}$ -enriched DON and DIN, although this only constituted a minor proportion of the total N leaching. The N leaching increased during autumn and winter due to an increase in DIN loss. Also the  $^{15}\text{N}$ -enrichment of lost N shifted from DON to DIN in the same period.

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# Nitrogen fixation and nitrogen allocation in red clover-grasslands

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## Abstract

Red clover (*Trifolium pratense* L.) is the dominant forage legume in Sweden. The <sup>15</sup>N natural abundance method and the <sup>15</sup>N isotope dilution method were used to measure N<sub>2</sub> fixation during different parts of the growing season in leys of three different ages. Measurements were made both in above-ground plant parts and in roots. The proportion of clover N derived from air, pNd<sub>fa</sub>, was consistently high in all parts of clover plants in all leys. Clover biomass, N content and N<sub>2</sub> fixation was consistently higher in late August (second cut) than in late June (first cut). Nitrogen fixation provided more than half of the total N removed in harvested biomass. Our efforts to study roots also highlight that both N<sub>2</sub> fixation and N allocated to plant parts below cutting height constitute large pools in the N budget for red clover grasslands. The considerable amounts of biomass and N that were left in roots and stubble after harvesting red clover-grass leys are important for regrowth of the plants and provide substantial N fertilization for the next crop in the crop rotation.

Keywords: ley age, N allocation above and below ground, <sup>15</sup>N methods, roots, seasonal variation in N<sub>2</sub> fixation, *Trifolium pratense*

## Introduction

Perennial grasslands for forage production, leys, cover a majority of cultivated land in Sweden. In northern Sweden the leys are typically sown with timothy, *Phleum pratense* L., and meadow fescue, *Festuca pratensis* L., together with red clover, *Trifolium pratense* L. These leys are commonly established as undersown crops in spring barley and are harvested in the next *ca.* 3 years. Two harvests per year for silage is common practice.

Red clover is of great value in forage consumption and milk production; it can improve nutrient uptake, especially divalent cations, from deeper soil layers, and because of its symbiosis with N<sub>2</sub>-fixing *Rhizobium leguminosarum* bv. *trifolii* red clover also has a great potential to increase sustainability in ley-dominated crop rotations (Frame *et al.*, 1998). Rhizobia infecting red clover showed a large genotypic variation but generally high N<sub>2</sub>-fixation capacity and are thus well adapted to the environmental conditions in north-Scandinavian soils so that no inoculation of clover is needed (Duodu *et al.*, 2007). In spite of the widespread use of red clover in Sweden, information about its N<sub>2</sub> fixation is scarce. In particular, there is a lack of information on N<sub>2</sub> fixation and consequent N allocation both above and below ground. Our work aimed to: 1) measure N<sub>2</sub> fixation in field-grown red clover, both as the proportion of N derived from N<sub>2</sub> fixation and as the amount of N<sub>2</sub> fixed on an area basis, in different plant parts including roots; 2) study seasonal variations and variations due to ley age regarding N<sub>2</sub> fixation in red clover; and 3) study the N budget in red clover grasslands of different ley age.

## Material and methods

We used the <sup>15</sup>N based ID (isotope dilution) and NA (natural abundance) methods to quantify the proportion of clover N derived from air (pNd<sub>fa</sub>) and the amount of N<sub>2</sub> fixed per area in a

first-year ley, a second-year ley and a third-year ley. The leys, with species composition as above, were in neighbouring fields in Umeå (63°49'N, 20°17'E), Sweden. Randomly distributed plots, 1 x 1 m, were fertilized with 0.67 g N m<sup>-2</sup> as <sup>15</sup>N-enriched (ID) or non-enriched (NA) KNO<sub>3</sub>. Biomass production, N content and N<sub>2</sub> fixation were studied separately in herbage cut at 5 cm, stubble and roots (down to 20 cm) in subplots 0.5 x 0.5 m from start of growing season until late June (first harvest) and late June until mid-August (second harvest) 1998. The grasses together with any weeds served as reference plants in both NA and ID methods. Further experimental details are given in Huss-Danell and Chaia (2005) and Huss-Danell *et al.* (2007).

## Results and discussion

In clover, the biomass production, N content and N<sub>2</sub> fixation per area was much higher at second harvest than at first harvest. Grasses, on the other hand, had more similar biomass production and N content at both harvests. pNd<sub>fa</sub> values in above-ground plant parts were very high, usually  $\geq 0.8$ . This is in agreement with findings that pNd<sub>fa</sub> in forage legumes grown in mixture with grasses in other temperate areas is high and is higher than in legume monocultures (reviewed by Carlsson and Huss-Danell, 2003). Nodulation and N<sub>2</sub> fixation in several African *Acacia* species was also increased by cultivating them together with grasses (Cramer *et al.*, 2007). It seems likely that a high competition for N by grasses is forcing legumes to high N<sub>2</sub> fixation in many ecosystems. Given that biomass is used to calculate N content (N concentration x biomass) in clover, and that pNd<sub>fa</sub> was fairly stable throughout our measurements, it follows that the amount of N<sub>2</sub> fixed per area was strongly dependent on clover biomass per area.

Table 1. N<sub>2</sub> fixation in herbage (above stubble) as proportion (%) of whole plants of red clover. Nd, not determined. Mean values  $\pm$  SD, n = 4. The difference between June and August was statistically significant for both methods in all leys (t-test,  $P < 0.05$ , Minitab 15 statistical software, Minitab Inc., PA, USA 2007).

Method, harvest	Ley 1	Ley 2	Ley 3
ID, first harvest (June)	49 $\pm$ 12	50 $\pm$ 13	41 $\pm$ 4.9
ID, second harvest (August)	79 $\pm$ 7.3	80 $\pm$ 8.7	81 $\pm$ 4.3
NA, first harvest (June)	Nd	51 $\pm$ 6.7	32 $\pm$ 6.5
NA, second harvest (August)	70 $\pm$ 2.7	79 $\pm$ 5.7	71 $\pm$ 13

Table 2. Nitrogen budget (g N m<sup>-2</sup>) for the growing season until second harvest, clover and grass together. The balance includes mineralization, residual N in stubble and roots as well as fixed N released to soil in previous year(s), N deposition, and any N losses to air and water. Total harvest was low, about 450 g DM m<sup>-2</sup>, this rainy year.

Measured	Ley 1	Ley 2	Ley 3
Input via N <sub>2</sub> fixation (whole plants)	5.24	5.40	7.52
Input via N fertilization	3.80	3.80	3.80
Removal (N in June + August harvests)	7.60	8.80	11.30
Remaining N in stubble + roots after August harvest	3.40	4.70	5.50
Calculated:			
Balance	1.96	4.30	5.48

Amounts of N<sub>2</sub> fixed measured in the herbage corresponded to about 45% of that in whole plants at first harvest but was considerably higher, about 77% of that in whole plants at the second harvest (Table 1). To include roots in field studies of N<sub>2</sub> fixation is a very demanding



work. The information presented here is therefore valuable for extrapolations of data from above ground to whole plants.

N<sub>2</sub> fixation played an important role in the N budget of the leys and corresponded to more than half of the N removed in harvest (Table 2). The amount of N remaining in stubble and roots increased with ley age. A part of this N is then used for regrowth of the plants after harvest and a part is decomposed in the soil. This 'carry-over' of N via remains in the field from ley 1 to ley 2 and from ley 2 to ley 3 is partly hidden in the last line in Table 2. The decomposition of remains is particularly important when the soil is ploughed for a next crop in the crop rotation.

## Conclusions

Our studies have highlighted that red clover consistently derives a majority of its N from N<sub>2</sub> fixation and have shown the need to consider plant parts below ground in N<sub>2</sub> fixation measurements as well as in N budgets in clover-grasslands.

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# Hay and silage as vitamin sources in organic sheep production

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## Abstract

The contribution of the essential vitamins A and E from organic forage-based rations was investigated, and how this affects the vitamin status of sheep. Two experiments were carried out during pregnancy and lactation in a herd consisting of 56 crossbred ewes. In the first study (I) hay and silage were compared. In the second study (II) two lambing times were compared and all the ewes had silage. Apart from forage, the rations contained barley, peas, rapeseed cake and minerals. There was no vitamin supplementation. Feed intake was registered and the vitamin content of feed and blood plasma was analysed.

Experiment I: Silage-fed ewes had higher levels of blood alfa-tocopherol in mid pregnancy (0.50 mg l<sup>-1</sup>) compared to hay-fed ewes (0.29 mg l<sup>-1</sup>). The intake of beta-carotene in the hay group was significantly lower than in the silage group. This was also reflected in blood vitamin A levels, which were higher in the silage group from the middle of pregnancy.

Experiment II: Early lambing ewes had higher blood levels of alfa-tocopherol during pregnancy compared with late lambers (in late pregnancy 0.91 and 0.58 mg l<sup>-1</sup>, respectively). One month after lambing, the late lambers were on pasture and had higher blood alfa-tocopherol levels (1.78 mg l<sup>-1</sup>) than the early group (0.81 mg l<sup>-1</sup>).

Keywords: sheep, silage, hay, alfa-tocopherol, vitamin A, lambing time

## Introduction

Vitamin A and E are essential parts of ruminant feed, and synthetic vitamins are included in most commercial concentrates and mineral feeds. In organic production systems, however, the use of synthetic additives is preferably avoided. Organic feed rations are mainly based on forage, which can contain an appreciable amount of vitamins. The forage vitamin content, therefore, has a large impact on the total vitamin intake of the animals. Hay normally contains less alfa-tocopherol (vitamin E) and beta-carotene (precursor of vitamin A) than silage (Hakkarainen and Pehrson, 1987). Pasture in general has a high content of beta-carotene and alfa-tocopherol, compared to conserved forages. This also clearly affects the levels of vitamins in the blood (e.g. Jukola *et al.*, 1996). The effect of grazing on vitamin A status of the animal lingers on into the housing season, due to storage in the liver. There is, however, very little storing of vitamin E in the body (McDonald *et al.*, 1995). The ewe's requirements of vitamins are also related to the stage of reproduction. The aims of this project were to study how the vitamin status in sheep was affected by feeding organic rations based on either hay or silage (Experiment I), and the relationship between lambing season and vitamin status (Experiment II).

## Materials and methods

The project was conducted at the Forage Research Centre in Umeå. The sheep flock consists of crossbreeds, mainly Swedish Landrace x Texel. The numbers of ewes were 25 and 31 in Experiment I and II, respectively. Both experiments started after housing at the end of

September. The feed rations were based on silage or hay, but barley, peas and rapeseed cake were also fed during pregnancy and lactation. Minerals were fed throughout the year, but no vitamins. All animals were kept in groups in straw-bedded pens during the experiments. Feed intake was registered two days per week in each pen. The feeds used were analysed for alfa-tocopherol and beta-carotene contents. Blood from the jugular vein was sampled at the start of the experiments, before mating, in mid and late pregnancy and once or twice after lambing, for analyses of alfa-tocopherol and retinol (vitamin A). One lamb from each litter was also blood sampled. The vitamin analyses were made at Research Centre Foulum at the University of Aarhus. In Experiment I, feed rations based on either hay or silage were compared. All ewes lambd in May/June. In Experiment II, two lambing times were compared, March/April and May/June. The feed rations were based on silage. Statistical analyses of individual blood vitamin levels were made with the programme NCSS 2000 (Hintze, 1999: [www.ncss.com/about\\_ncss.html](http://www.ncss.com/about_ncss.html)). SAS (Ver. 8.02, SAS Institute Inc., Cary, NC, USA) was used for calculations on intake per pen.

## Results and discussion

The content of beta-carotene in hay and silage was on average 8 and 23 mg kg<sup>-1</sup> DM, respectively. There was almost no beta-carotene in the concentrates. Hay and silage contained 8 and 19 mg kg<sup>-1</sup> DM alfa-tocopherol, respectively. Barley and rapeseed cake contained 50 and 108 mg kg<sup>-1</sup> DM alfa-tocopherol, respectively. Table 1 shows the average daily intake of vitamins in the last eight weeks of pregnancy. The recommended intake of vitamin A the last month before lambing is 3,640 IU per day (National Research Council, 2007). According to this, all treatments seem to have had an adequate intake, although the hay group had a lower intake than the others. The intake of vitamin E was far below the recommended 448 IU per day (National Research Council, 2007) on all treatments.

Table 1. Mean intake of vitamins per ewe and day in the eight last weeks of pregnancy, means per treatment in experiments I and II (standard deviation in brackets).

	Experiment I			Experiment II		
	Hay group	Silage group	Sig.	Early lambing	Late lambing	Sig.
Vitamin A, IU <sup>1</sup>	4,750 (110)	27,590 (1,973)	$P < 0.01$	9,150 (155)	6,920 (56)	$P < 0.01$
Vitamin E, IU <sup>2</sup>	36 (0.3)	34 (1.5)	ns	44 (0.4)	38 (0.2)	$P < 0.01$

<sup>1</sup>1 mg beta-carotene = 671 IU vitamin A.

<sup>2</sup>1 mg plant-derived alfa-tocopherol = 1.49 IU vitamin E. 1 mg gamma-tocopherol = 0.15 IU vitamin E.

Mean values of retinol in the blood plasma in Experiment I are shown in Figure 1. The level was higher in the silage group from the middle of pregnancy and onwards. There also was a significant difference when comparing blood from their one-week-old lambs. In Experiment II the treatments differed in blood retinol levels only in the middle of pregnancy, when the plasma from the early lambing ewes contained 0.26 mg retinol l<sup>-1</sup> and that from the late lambers contained 0.21 mg l<sup>-1</sup> ( $P < 0.001$ ). The reason why there was no difference in late pregnancy is probably because the intake of vitamin A was above the recommended levels on both treatments and the difference between them was not so big. At one month of age the late-born lambs were on pasture and had significantly higher average level of plasma retinol, than the early born lambs that still were indoors at that age (0.34 mg l<sup>-1</sup> vs. 0.28 mg l<sup>-1</sup>,  $P < 0.05$ ).

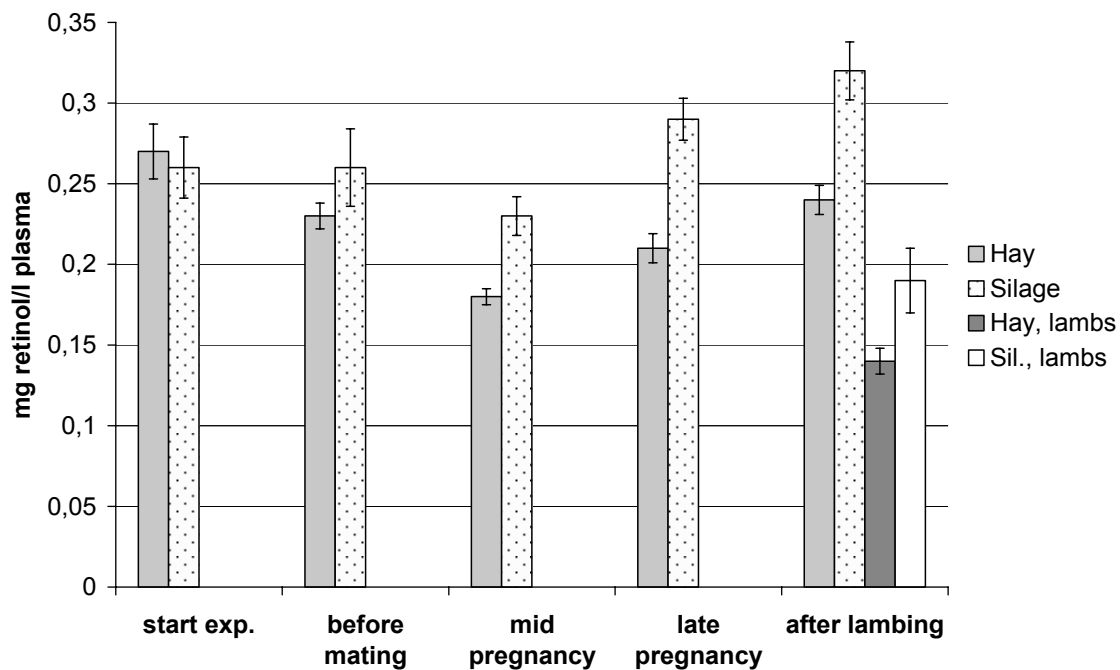


Figure 1. Retinol (+ / - standard error) in blood plasma from ewes and lambs in Experiment I.

The only significant difference in plasma alfa-tocopherol in Experiment I was in mid pregnancy, when the average level in the silage group was  $0.50 \text{ mg l}^{-1}$  and in the hay group  $0.29 \text{ mg l}^{-1}$  ( $P < 0.001$ ). The rapeseed that was fed to all ewes in late pregnancy and in lactation probably evened out the differences at that time. In Experiment II the early lambing ewes had significantly higher plasma alfa-tocopherol levels than the late lambers from the start until late pregnancy (at mating  $1.35$  and  $0.62 \text{ mg l}^{-1}$  in the early and late-lambing groups, respectively,  $P < 0.001$ ; in mid pregnancy  $0.82$  and  $0.68 \text{ mg l}^{-1}$ ,  $P < 0.05$ ; in late pregnancy  $0.91$  and  $0.58 \text{ mg l}^{-1}$ ,  $P < 0.001$ ). One month after lambing, when the late-lambing group was on pasture, both ewes and lambs had higher plasma-alfa-tocopherol levels than in the early group (ewes  $0.81$  and  $1.78 \text{ mg l}^{-1}$  in the early and late-lambing groups, respectively,  $P < 0.001$ ; lambs  $0.51$  and  $1.16 \text{ mg l}^{-1}$ ,  $P < 0.001$ ).

## Conclusions

In accordance with the vitamin content in the feed, silage-based feed rations give higher plasma concentrations of vitamins, especially retinol, compared with rations based on hay. Lambing early in the season results in higher plasma-vitamin levels during pregnancy. Later lambing ewes go onto pasture soon after lambing and get a higher vitamin intake during lactation.

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# Analysis of hyperspectral data to estimate dry matter yield of legume-grass swards

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## Abstract

To examine the potential of field spectral measurements for a non-destructive assessment of dry matter yield (DM) from legume-grass swards, a pot experiment was conducted at the University of Kassel in Germany. The plants were sown in binary legume-grass mixtures and pure swards composed of red clover (*Trifolium pratense* L.), white clover (*Trifolium repens* L.), lucerne (*Medicago sativa* L.) and perennial ryegrass (*Lolium perenne* L.). Three days before harvesting the reflection of the incident light on the swards was measured with a spectrometer (FieldSpec, Analytical Spectral Devices) in the range 350-2,500 nm. For further data processing, enhanced vegetation index (EVI) and partial least square regressions (PLS) were calculated and related to the DM yield of each sward. Both methods estimated DM yields with high coefficients of determination (0.90 and 0.94), whereas the PLS produced slightly better results than the EVI.

Keywords: Legume-grass swards, dry matter yield, enhanced vegetation index, hyper spectral analysis

## Introduction

The growth of legume-grass swards is affected by several parameters such as soil texture, water availability, nutrient supply, temperature and solar insolation (Ledgard and Steele, 1992). The goal is to establish an efficient technique to assess information on plant biomass without disturbing the growth of swards. Such methodology would help to better understand growth and nitrogen dynamics, and thus enable a more accurate management of the swards (Ehlert and Adamek, 2005; He *et al.*, 2007).

## Materials and methods

Ten different experimental swards were investigated in a greenhouse in the year 2004/2005 at the University of Kassel in Germany. Beside pure swards of perennial ryegrass (*Lolium perenne* L.) (seed rate 20 kg ha<sup>-1</sup>), red clover (*Trifolium pratense* L.) (8 kg ha<sup>-1</sup>), white clover (*Trifolium repens* L.) (4 kg ha<sup>-1</sup>) and lucerne (*Medicago sativa* L.) (16 kg ha<sup>-1</sup>) the following mixtures were sown: red clover (8 kg ha<sup>-1</sup>) with perennial ryegrass (20 kg ha<sup>-1</sup>) (R8P); red clover (2 kg ha<sup>-1</sup>) with perennial ryegrass (20 kg ha<sup>-1</sup>) (R2P); white clover (4 kg ha<sup>-1</sup>) with perennial ryegrass (20 kg ha<sup>-1</sup>) (WP); and lucerne (16 kg ha<sup>-1</sup>) with perennial ryegrass (20 kg ha<sup>-1</sup>) (LP).

To compare swards at different growth stages the legume-grass mixtures were sown every two weeks and were all harvested on the same dates (21, 35, 49 and 63 days after sowing). The sowing was carried out manually with a distance between rows of 12 cm and at a sowing depth of 0.5 cm. The pots (70 x 70 x 20 cm) were filled with 2 cm drainage substratum (Lavagrus) and about 16 cm homogenized loamy soil (sL- 1S; 3.6% sand, 73% silt, 23.4% clay and 2% humus). Soil analysis indicated optimum levels of phosphorus, magnesium and potassium and a pH-value of 6.7. No fertilizers were used. To determine the content of grass,

legumes and unsown plants in the sward, total above-ground biomass was separated in its fractions. After the determination of all species the samples were dried at 65 °C for 24h. Three days before harvesting, all swards were measured under artificial light conditions (quartz tungsten halogen lamp; JVC 14.5V-50W C) using a spectrometer (FieldSpec, Analytical Spectral Devices) which measured the reflection of the incident light on the swards in the range between 350 and 2,500 nm. To reduce the amount of spectral data points the range of 350 to 1,000 was compressed to intervals of 10 nm and the range of 1,000 to 2,500 to intervals of 30 nm respectively using the resampling procedure of the program ENVI (4.1). With the resampled data the enhanced vegetation index (EVI) was calculated (Huete *et al.*, 2002) and then related to the DM yield of the swards; (M = Mean):

$$EVI = 2.5 * \left( \frac{(M_{800nm} - 900nm) - (M_{650nm} - 700nm)}{(M_{800nm} - 900nm) + 6 * (M_{650nm} - 700nm) - 7.5 * (450nm - 500nm) + 1} \right)$$

In addition, a partial least square (PLS) regression was calculated with the whole resampled dataset and the DM yield using the program Grams/AI<sup>tm</sup> (Thermo Galactic, USA).

## Results and discussion

All examined swards were still in the vegetative stage of tillering. Swards grown for 63 days after sowing had an almost closed canopy (BBCH stage 29 for grass and BBCH stage 22 for legumes; Meier, 2001). The 49-day-old swards were at the beginning of row closure (BBCH 25 for grass and BBCH 19 for legumes) whereas the 35- and 21-day-old swards were still very small. While the grass of the 35-day-old swards already began to tiller (BBCH 19/20) and the legumes developed their third or fourth leaf (BBCH 13/14) the grass and legumes of the youngest swards were still developing their first leaves (BBCH 12/13 and BBCH 10 respectively).

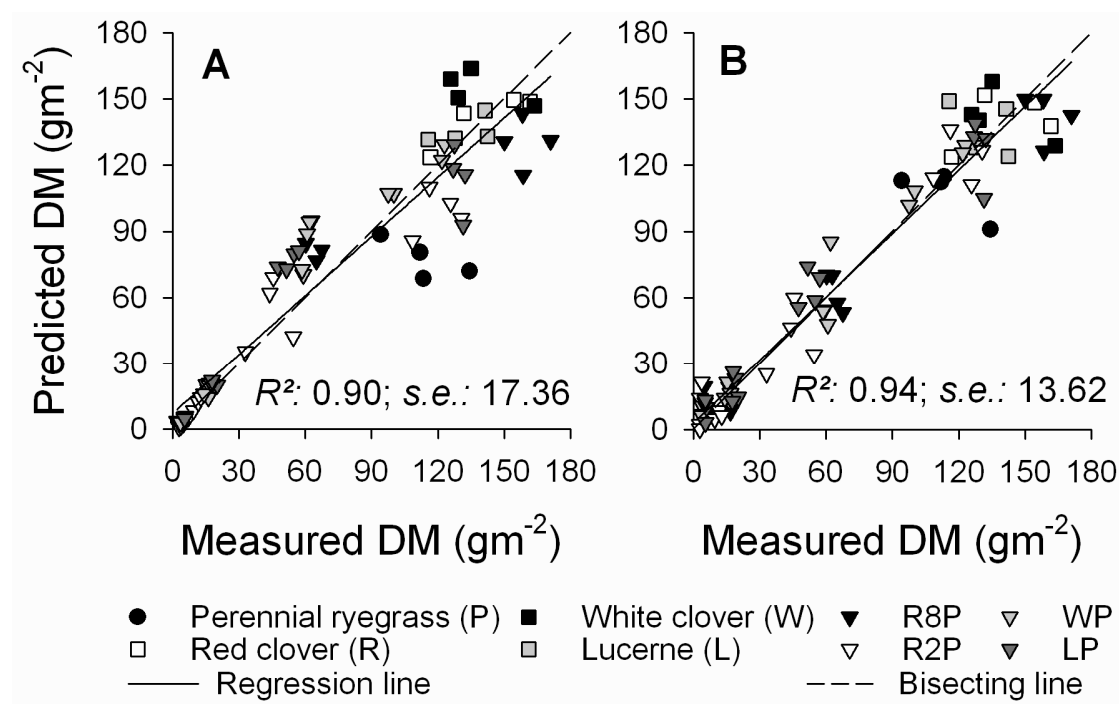


Figure 1: Measured and predicted DM yield (gm<sup>-2</sup>) of different legume-grass swards by the vegetation index EVI (A) and the PLS regression (B) respectively.

The DM proportion of legumes in the experimental swards had a maximum in the pure swards of 0.69 and a minimum in the mixtures at 0.96. In contrast, the proportion of grass was always higher yielding to a maximum of 0.792 in the pure swards and to a minimum of 0.454 in the mixtures.

As swards were investigated at different stages of development, total above-ground DM yields varied from 3 g m<sup>-2</sup> for the youngest swards to 159 g m<sup>-2</sup> to the oldest swards. The differences in total yield were mainly caused by the legumes, as the yield of the grass did not differ much among treatments. Additional variation in yield and contribution of legumes was induced by varying the seed rate of red clover between 8 kg ha<sup>-1</sup> (R8P) and 2 kg ha<sup>-1</sup> red clover (R2P).

Estimation of the DM yield through the vegetation index EVI and the PLS regression showed good results with coefficients of determination of 0.9 for EVI and 0.94 for PLS, respectively (Figure 1). However, EVI underestimated the DM yield more than the PLS regression. In particular, the pure swards of perennial ryegrass were underestimated by EVI, whereas pure swards of white clover and legume-grass mixtures with DM yields between 60 to 90 gm<sup>-2</sup>, were overestimated. In contrast, the DM yield detection by the PLS regression did not show these tendencies, resulting in a lower standard error of 13.63 gm<sup>-2</sup> in comparison with the EVI (17.36 gm<sup>-2</sup>).

Despite these differences, both methods seem to be suitable for the DM yield detection. Particularly the EVI would be preferable in practice as its calculation needs only a small range of the whole reflection spectrum and already existing sensors like the YARA N-sensor<sup>®</sup> could be applied.

As these results were obtained under controlled conditions in a greenhouse, the next step would be to examine the spectral characteristics of legume-grass swards in the field. Furthermore, not only vegetative but also generative growth stages should be investigated. If the results can be confirmed also under field conditions, the EVI could be an efficient tool for the non-destructive assessment of DM yield in the field.

## Conclusions

The results of this experiment show that the DM yield of legume-grass mixtures can be detected through both approaches of the EVI and the PLS regression. Altogether, the hyperspectral data analyses of the PLS showed slightly better results than the estimation of DM yield through the EVI. Nevertheless, for an implementation of spectral measurements in practice the EVI would be more suitable as its calculation needs only a small range of the whole reflection spectrum. Further research is needed to verify the results under field conditions and at generative growth stages.

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# **Influence of organic fertilizers on the dry matter yield and microelement content of meadow herbage**

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## **Abstract**

In a field experiment, the effects of organic manures (farmyard manure and vermicompost), with treatments alone and in combination with NPK fertilizer, were studied in terms of their effects on herbage dry matter yield and microelement concentrations (Mg, Cu and Zn) in the sward. The results showed that the organic manure treatments, and especially the farmyard manure in combination with NPK mineral fertilizers, increased the dry matter yield of the meadow. Changes in the fodder quality of the meadow sward, in terms of microelement contents, were detected. The highest Mn concentrations were in herbage from the meadow sward fertilized with vermicompost, but concentrations of Cu and Zn were higher in the sward fertilized with farmyard manure.

**Keywords:** farmyard manure, vermicompost, grassland, microelements content

## **Introduction**

Meadows are commonly fertilized with mineral fertilizers. However, many authors (Jankowska-Huflejt, 2000) have reported that mineral fertilizers can cause undesirable changes in the botanical and chemical composition of the meadow sward. In these situations farmers should also consider the use of organic materials (Baran and Martyn, 1996; Lekman *et al.*, 1997). The aim of this work was to investigate the possibility of utilization of different organic manures, alone and in combination with NPK fertilizers, on the production and fodder quality of meadow grassland.

## **Materials and methods**

A randomized block experiment with four replicates of each treatment was carried out over 3 years (1999-2001). The area of each plot was 1.5 x 6 m. Treatments consisted of the following fertilizer combinations:

1. Control
2. NPK (as mineral fertilizer)
3. Farmyard manure
4. Farmyard manure + NPK
5. Vermicompost
6. Vermicompost + NPK

All the organic manures were applied once in spring 1999 at the rate of 10 t organic matter per hectare. The mineral fertilizer was applied each year, but nitrogen ( $180 \text{ kg N ha}^{-1}$ ) and potassium ( $125 \text{ kg K ha}^{-1}$ ) were applied in three split dressings. The phosphorus fertilization ( $48 \text{ kg P ha}^{-1}$ ) was applied once in the spring each year. During the growing season each year three cuts were taken. The fresh material from each plot was dried and weighed separately to calculate herbage dry matter (DM) yield. In addition a 1 kg sample was collected at each cut from each plot for chemical analysis. For comparison of differences between the means the Tukey test was used to determine significance at the level of  $P \leq 0.05$ .

## Results and discussion

Chemical composition of the organic manures is summarized in Table 1. Forage yield and quality of the sward were substantially affected by different kind of fertilizers (Table 2).

Table 1. Chemical composition of farmyard manure and vermicompost.

Fertilizer	Organic matter	% DM	pH	Major nutrient concentrations in % DM						Micro-nutrient concentrations in mg kg <sup>-1</sup>				
				N	P	K	Ca	Mg	Na	Zn	Cu	Cd	Pb	Cr
Manure	73.5	21.0	7.0	2.51	0.48	1.67	1.38	0.53	0.11	178	41	0.6	9.0	7.0
Vermicompost	52.9	43.9	7.0	1.37	1.05	1.68	0.17	0.04	0.01	220	20	0.62	9.0	7.0

Combined organic and mineral fertilization, especially farmyard manure + NPK (3-year mean 10.10 t DM ha<sup>-1</sup>), or vermicompost + NPK (3-year mean 9.02 t DM ha<sup>-1</sup>) had the most positive influence on DM yields, irrespective of years (Table 2).

Table 2. Yield of dry matter in t ha<sup>-1</sup> in relation to fertilizer treatment

Treatment	Year of study			Mean
	1999	2000	2001	
1. Control	3.88	3.75	3.31	3.65
2. NPK fertilization	7.01	7.50	9.57	8.03
3. Farmyard manure	6.37	7.55	8.13	7.35
4. Farmyard manure + NPK	9.10	9.55	11.65	10.10
5. Vermicompost	6.03	5.83	7.12	6.32
6. Vermicompost + NPK	7.96	8.28	10.82	9.02
LSD for treatment	0.14			
for years	0.05			
for interaction treatment x years	0.24			

The DM yields of the meadow for the treatments fertilized separately with different kinds of organic materials (treatments 3 and 5) were about 3 t DM ha<sup>-1</sup> greater than the unfertilized control, but were lower by about 3 t ha<sup>-1</sup> compared with the corresponding treatments which received NPK fertilizer in addition (treatments 4 and 6). The microelement concentrations in the harvested herbage also depended on the fertilization treatment. Content of manganese (Mn) (Table 3) was generally optimal. The highest content of manganese (76.36 mg Mn kg<sup>-1</sup>) was in fodder from plots fertilized with vermicompost together with NPK (treatment 6). For the other microelements, the highest concentrations of copper (Cu) and zinc (Zn) were from the sward fertilized with farmyard manure + NPK (treatment 4) and amounted to 10.85 and 16.99 mg kg<sup>-1</sup> of Cu and Zn, respectively.

## Conclusions

The vermicompost treatment resulted in a similar effect on DM yield as the farmyard manure treatment. The highest manganese concentrations were in herbage from the meadow sward fertilized with vermicompost, but concentrations of copper and zinc were higher in meadow sward fertilized with farmyard manure than with vermicompost. This work has show that there are good prospects for the use of unconventional organic fertilizers (e.g. vermicompost) for meadows.

Table 3. Herbage concentrations of manganese, copper and zinc (mg kg<sup>-1</sup> DM) in fodder from meadow fertilized by different fertilizers and combinations in 1999- 2001.

Meadow fertilized by different fertilizers and combinations in 1999-2001.				
Treatment	Years of study			Mean
	1999	2000	2001	
Manganese				
Control	43.91	47.91	37.42	43.08
NPK fertilization	66.75	90.67	47.67	68.36
Farmyard manure	56.38	69.75	52.67	59.60
Farmyard manure + NPK	64.58	59.58	77.67	67.28
Vermicompost	44.75	53.58	43.50	47.28
Vermicompost + NPK	72.50	75.00	81.58	76.36
LSD for treatment 0.06; for year 0.2; treatment x year 1.1				
Copper				
Control	12.13	9.60	9.25	10.38
NPK fertilization	12.33	10.91	9.16	10.80
Farmyard manure	11.66	11.00	8.33	10.33
Farmyard manure + NPK	13.16	9.66	9.75	10.85
Vermicompost	11.66	9.00	8.91	9.86
Vermicompost + NPK	10.33	11.25	10.58	10.72
LSD for treatment ns; for year 0.2; treatment x year n.i.				
Zinc				
Control	10.45	7.65	9.76	9.76
NPK fertilization	13.40	16.79	11.69	11.69
Farmyard manure	12.45	7.79	12.50	12.50
Farmyard manure + NPK	23.15	8.86	16.99	16.99
Vermicompost	13.58	7.81	7.38	7.38
Vermicompost + NPK	15.51	14.47	11.40	11.40
LSD for treatment 0.14; for year 0.05; treatment x year 0.24				

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# Establishing a risk index to prevent nitrogen non-point pollution of waters in the Basque Country

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## Abstract

The intensification of the livestock sector in the Basque Country has led to environmental impacts in the soil-plant-atmosphere system. The necessity of protecting natural resources has promoted the development of new spatial tools for identifying and classifying the non-point pollution risk areas where preventive measures should be applied. We have adapted runoff indexes and leaching indexes to specific characteristics for the Basque Country and available data sources. The adapted indexes have been applied on the Gipuzkoa pasture area; as a result 32% of total pastures have been classified as having a high potential risk of discharging N into water courses. This location of the pastures and farms with high potential risk to pollute surrounding waters represents the first approach in the development of a subsequent *in situ* study to improve pasture management in Gipuzkoa.

Keywords: Nitrogen, non-point pollution, runoff index, leaching index, manure and slurry

## Introduction

The intensification of the livestock sector in the Basque Country is the origin of environmental impacts in the soil-plant-atmosphere system. The necessity of protecting natural resources has led to the implementation of new spatial tools that identify and classify the non-point pollution-risk areas where preventive measures should be applied. The aim of these tools is to locate fields that present high potential risk of pollution, and also to identify the most important management practices affecting pollution processes. Initially, most emphasis has been placed on nitrogen (N) because of the high solubility of nitrate and its associated potential problems for human health. The objectives of this study are (i) to adapt leaching and runoff indexes of N to a specific area of the Basque Country for classifying pasture surfaces in terms of different potential for pollution, and (ii) to characterize, based on the previous classification, the type of farms presenting high risk to export N to water resources, for the subsequent development of *in-situ* detailed studies.

## Materials and methods

All the bovine, ovine, goat or equine farms of Gipuzkoa (3,948 farms) and their associated pastures (26,759 ha) were identified. For this area, two N indexes (Runoff Index and Leaching Index) reflecting the potential for N export to water resources were calculated and applied. These indexes have been adapted from indexes developed by other authors (Heathwaite *et al.*, 2000; Sharpley *et al.*, 2003). Each index includes transport factors which are related to the specific site characteristics such as slope, texture, distance to water courses (Tables 1 and 2). Each transport factor has a numerical value assigned, indicating its relative importance in contributing to N export: none (0), low (1), medium (2), high (4) or very high (8). Each index is calculated by adding the values of all factors.

The spatial data needed to calculate the indexes were obtained from the Basque Government: land use, lithology, permeability, organic matter in soils, aquifer location (1:25,000), water

courses (1:5,000) and topography (1:10,000), and this information was incorporated into a geographic information system (ArcGIS 9.2). In this way, the runoff index and the leaching index are calculated and these are integrated into a unique N Transport Index (NTI), assuming that the value of the NTI is the most limiting of both transport indexes. The NTI is considered a measure of the effectiveness of this site for transporting N that is potentially available from various sources.

Table 1. Runoff transport factors (Runoff TF), assigned values and their interpretation.

Runoff TF	None (0)	Low (1)	Medium (2)	High (4)	Very high (8)
Distance to water courses	> 170 m	130-170 m	80-130 m	30-80 m	< 30 m
<sup>1</sup> Runoff Class	0	1	2	4	8
<sup>2</sup> Erodibility (K factor)	≤ 26	27-31	32-34	35-37	38-39
Sum of N Transport Factors		0-2	3-8	9-14	> 14
Runoff Index interpretation		Low	Medium	High	Very high

<sup>1</sup>The runoff class involved the slope, hydrology group, land use and vegetation cover.

<sup>2</sup>The erodibility factor (K) of the Universal Soil Loss Equation (USLE) was calculated using Knisel (1992) simplification.

Table 2. Leaching transport factors (Leaching TF), assigned values and their interpretation.

Leaching TF	None (0)	Low (1)	Medium (2)	High (4)	Very high (8)
Texture		Clay, silty clay	Clay loam, silty clay loam, sandy clay	Sandy loam, loam, Silt loam, sandy clay loam	Sand, sandy loam
Permeability		Impermeable	Low	Medium	High
Distance to aquifers	> 170 m	130-170 m	80-130 m	30-80 m	< 30 m
Slope		> 15%	8-15%	5-8%	< 5%
Sum of N Transport Factors		0-10	11-16	17-22	> 22
Leaching Index interpretation		Low	Medium	High	Very high

Finally, NTI was combined with management characteristics related to the amount of N in soil (Source Factors) resulting in the N Global Index (NGI), which was an estimate, or indicator, of available N that was likely to be transported off the field (Table 3).

Table 3. Nitrogen Global Index based on N Transport Index and N Source Factors.

N Transport Index	N Source Factor (kg N ha <sup>-1</sup> yr <sup>-1</sup> )	N Global Index
High-Very High	None (0)	Low
Low-Medium	Low (0-170)	Low
High-Very High	Low (0-170)	Medium
Low-Medium	Medium (170-300)	Medium
High-Very High	Medium (170-300)	High
Low-Medium	High (> 300)	High
High-Very High	High (> 300)	Very High

The Source Factor considered is the sum of the amounts of organic and mineral N applied. The organic N applied is estimated depending on the number and type of animals in each farm and the distances from the farm to the pasture fields, assuming that the longer distance from farm to field, the lower quantity of organic N is applied. On the other hand, no mineral N is applied on pastures located at > 600 m elevation, or on pastures with a slope of more than 20%, while it is assumed that an average of 40 kg N ha<sup>-1</sup> year<sup>-1</sup> is applied on the rest of pastures. The information about livestock (number, type and N excretion by type), manure management systems of each farm, and mineral fertilization rates were obtained from the Basque Government Statistical Service.

## Results and discussion

The potential for N export from the field, due to the combination of transport and source factors (Table 4), is high or very high at 31% of the total pasture area (8,443 ha) because the organic and mineral fertilization rate is higher than  $170 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ ; and, in some cases (6,228 ha), that fact is combined with a high or very high transport risk. For 35% of the pasture area the N-export potential risk is low, with the low risk being due to transport factors and, in some cases, to low N-application rates ( $< 170 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  or even none). Finally, the N-export potential is medium for 34% of the area. Part of this pasture area (3,084 ha) is slightly over-fertilized ( $170\text{-}300 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ ) but the N-export potential due to transport factors is low or medium, and the other part of such pastures (5,979 ha) are not over-fertilized ( $< 170 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ ) but the N risk due to transport factors index is high or very high.

Table 4. Pasture area with different potential to export N to water courses (NGI).

NGI	Area (ha)	Percentage (%)
Low	9253	34.6
Medium	9063	33.9
High-very high	8443	31.5
Total	26759	100

The farms having more than 6 ha in the high-to-very high categories of N export vulnerability were grouped and considered the most important farms to reduce the N export to water courses in the province of Gipuzkoa. The 407 farms included in this group generate 44% of the total N excreted in Gipuzkoa and they amount to 56% of the total area included in the high-to-very high categories of N-export potential. Moreover, the average stocking rate exceeds the equivalent of  $170 \text{ kg N ha}^{-1} \text{ yr}^{-1}$  in 87% of such farms, suggesting potential problems in the management of the slurry and manure in accordance with the established limit in the Directive 91/676/EEC. More than half of the N generated in these 407 farms (53%) was excreted by dairy cattle, while beef cattle and sheep excreted 14% and 17%, respectively.

## Conclusions

The potential for N export to water courses was high or very high in 31% of the pasture area of the province of Gipuzkoa, so manure and slurry management in this area will need preventive agricultural practices. The improvement of slurry management on the farms of Gipuzkoa should, first, be focused on dairy cattle farms, because most of the pasture area having high potential for N-export to water courses was located on such farms and most of the N excretion was generated by dairy cows. Nevertheless, the pastures in Gipuzkoa should be studied *in situ* to validate statistically the results based on the NGI, beginning with the 407 farms considered as the most important sources of N to waters and to determine the adequacy of the legally established N application limits.

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# Milk urea content: effects of environmental parameters and relationships with other milk traits

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## Abstract

Milk urea content can be an indicator of dietary N imbalance and a useful tool for ration planning to reduce N waste and feed costs. In Belgium, urea concentration in milk from commercial dairy farms is routinely determined along with the official milk analyses carried out on milk samples for the dairies. Thus, 5,548,174 records obtained over 7 years were analysed using a GLM model including the effects of year, month, somatic cell count, fat and protein concentration in milk, and different agricultural areas in Wallonia distinguished by soil, crop and cattle diet. The interactions between years and areas and between months and areas were also included in the analyses. The agricultural areas influenced strongly the milk urea content, according to feed practices derived for sugar beet and cereals production or grassland management. Milk urea content was significantly correlated to milk protein content:  $\text{urea (mg l}^{-1}\text{)} = -139 + 11.89 \text{ protein content (g l}^{-1}\text{)}$ . Milk urea levels changed also largely according to the months, reflecting changes in diet composition. Interaction between agricultural areas and month also had a significant influence on milk urea content, while somatic cell count, milk fat content and year effects had less impact.

Keywords : milk, urea, agricultural area, month, protein, diet

## Introduction

Environment, nitrogen (N) losses and pollution are acute problems, and scientists of many countries are involved in estimations of nitrogen losses from cattle production with the aim of decreasing these losses. Accurate estimates of N excretion are needed to plan manure storage facilities and to manage nutrient use. Urea is a protein degradation waste which is related to N intake. In Belgium, milk urea content is easily and routinely estimated in tank milk and is a useful parameter to assess the balance of the diet for grazing dairy cows (Geerts *et al.*, 2004). Knowledge of milk urea content can be useful in the management of dairy cows. This study aims to evaluate for Wallonia (southern Belgium) the influence of the year, agricultural area, month, somatic cell count, protein and fat content on milk urea content over a 7-year period.

## Materials and methods

Milk samples from tanks obtained when the milk was transferred from the farms to the dairies were analysed by the Comité du Lait. Urea, fat and, protein concentrations were determined by infrared method (Milkoscan FT 6000<sup>®</sup> – Foss Denmark). On average there were 12 samples per month and per milk producer, so for Wallonia there were 5,548,174 records obtained over 7 years, from 2000 to 2006. Data were analysed using a GLM model including the effects of year, month, somatic cell count (SCC), fat and protein concentration in milk and agricultural areas in Wallonia distinguished by soil, crop and cattle diet. The interactions between years and areas and between months and areas were also included in the analyses. The agricultural areas were Ardennes, Condroz, Fagne, Famenne, Haute Ardenne, Herbagere, Jurassique, Limoneux and Sablo-limoneux.



## Results and discussion

The average yearly milk urea content was 262.5 mg l<sup>-1</sup>. Values between 150-350 mg l<sup>-1</sup> can be considered as normal. There were significant effects for all variables ( $P < 0.001$ ). The statistical model explained 38% of the variation. The part of the variation was 41.2%, 19.1%, 17.7%, 16.0% for the agricultural area, protein concentration, month and area-month interaction, respectively. The effects of year, year-area interaction, milk fat concentration and SCC, explained 2.3%, 1.6%, 1.6% and 0.3%, respectively, of the variation. Meura *et al.* (2007) analysed data from one year (2005) and explained 31% of the variation with the following variables in the model: fat concentration, protein concentration, month, agricultural area and area-month interaction. The part of the variation was higher for month and lower for area-month interaction in the present study.

The lowest milk urea contents were found in the areas of Sablo-limoneux and Limoneux (228.7 and 232.6 mg l<sup>-1</sup>), and the highest in the Ardennes, Herbagere and Haute Ardenne areas (292.2, 290.3, 295.0 mg l<sup>-1</sup>; Table 1). Jurassique, Famenne and Condroz showed intermediate concentrations (257.9, 268.4, 263.3 mg l<sup>-1</sup> respectively). There were major differences in agricultural practices in these areas. Limoneux and Sablo-limoneux, with 32.8% of the total number of cows in rather small size herds were characterized by the production of crops which were mainly cereals and beets. By contrast, in the Ardennes, Herbagère and Haute-Ardenne areas, grass was the main crop.

Table 1. Main crops and characteristics of the dairy farms according to the agricultural areas in 2005.

Agricultural areas	Main crops	Cow number		Farm <sup>-1</sup>	Mean milk urea content mg l <sup>-1</sup>
		Total	Proportion of total (as %)		
Haute Ardenne	Pasture	24037	10.3	41	295.0
Ardennes	Pasture	23577	10.2	34	292.2
Herbagère	Pasture	50243	21.7	52	290.3
Famenne	Pasture	19072	8.2	41	268.4
Condroz	Pasture, cereals, beet	31726	13.7	29	263.3
Jurassique	Pasture	7534	3.2	43	257.9
Limoneux	Cereals, beet, pasture	64529	27.8	31	232.6
Sablo-Limoneux	Cereals, beet, pasture	11492	5.0	32	228.7

Months explained 17.7% of the variation of the milk urea content. Urea content showed large seasonal variations. Milk urea content was the highest in August (356.4 mg l<sup>-1</sup>) and the lowest in January (200.1 mg l<sup>-1</sup>; Figure 1). From November to March, urea content was lower than during the April to June period. In July, August, September and October, urea content showed the highest concentration of the year. The overall low urea content evolution from November to March can be related to the indoor management of the cows. On the basis of the grazing period from April to October, one has to note that the urea content was intermediate in April, May, June, and then rose to high levels up to October. Indoor period and grazing period were different in terms of feeding management. During the outdoor period, dairy cows generally grazed on most farms and their diet consisted mainly of grass containing a large excess of protein relative to the energy content. By contrast, during the indoor winter period, the diet was generally more diversified and balanced. During the grazing period, average milk urea content in May and June (282 mg kg<sup>-1</sup>) was lower than in July, August and September (343 mg kg<sup>-1</sup>). The main changes in grass composition were an increase in nitrogen content

and a decrease in water soluble carbohydrates content during the grazing season. Such changes in chemical composition can lead to a greater imbalance of the diet at the end of the grazing season and, as a result, higher milk urea contents during this period.

Milk protein content explained 19.1% of the variation of the milk urea content according to the significant relationship ( $P < 0.001$ ):  $\text{urea (mg l}^{-1}\text{)} = -139 + 11.89 \text{ protein content (g l}^{-1}\text{)}$ .

Interaction between areas and months explained 16.0% of the variation in concentration of milk urea ( $P < 0.001$ ). In each area, milk urea content increased sharply from March to July. The pattern over the year of milk urea content in the Ardennes was quite different compared with the Haute Ardennes and Herbagère, the highest milk urea content being observed during the grazing period and intermediate milk urea content during indoor period. In the Ardennes no supplement was offered to cows when they grazed, while during the indoor period the cows were offered mainly hay and grass silage of rather poor quality owing to a late harvest. The management in the Ardennes can be considered as being relatively extensive compared with the management in the Herbagère and Haute-Ardenne areas. In these areas, the dairy herd was usually larger and milk was often the only production on the farm, pastures being intensively managed and supplemental feed being distributed during the grazing season.

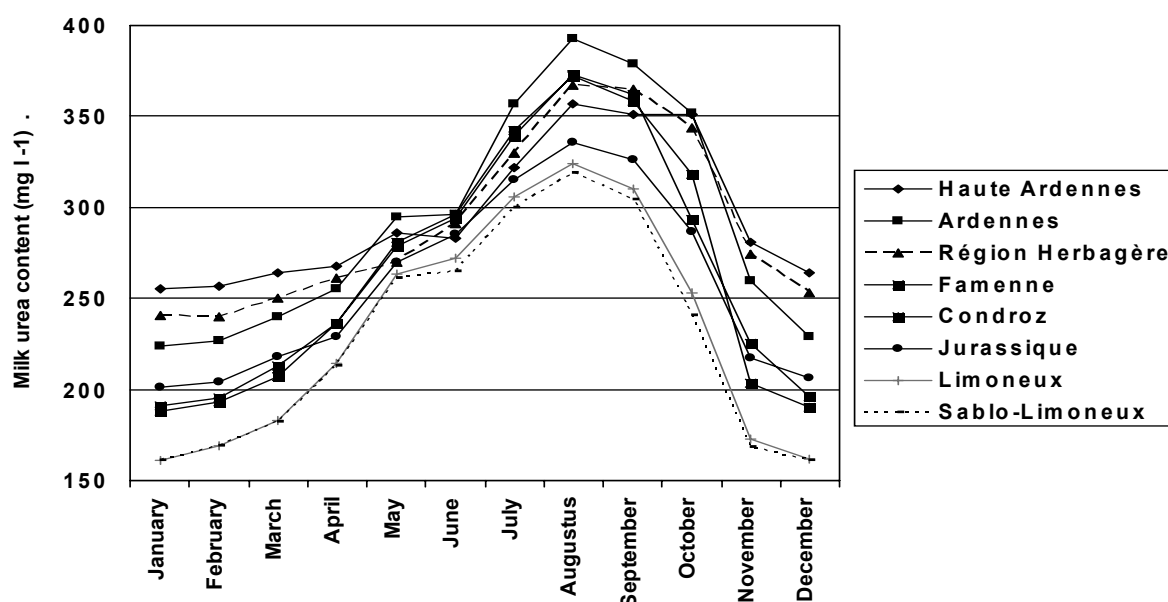


Figure 1. Monthly milk urea content in different agricultural areas.

## Conclusions

Agricultural areas, milk protein contents and months explained the variation in milk urea contents. Highest values were found during the grazing period and in areas where grass was the main crop. Nitrogen intake can explain the differences observed in the agricultural areas and during the year. Other dietary components, such as carbohydrate fractions, could also influence milk urea content. More research is needed on the effects of dietary components.

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# Strategy of organic fertilizer use on permanent grassland – results of a 22-year-old experiment on meadow and mowing-pasture

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## Abstract

In a 22-year-old experiment in Southwest Germany, the effects of different fertilization systems (organic and mineral fertilizers) on permanent grassland were investigated. The effects were compared under meadow use, as well as for a mown pasture with two grazings per year. Field plots were 25 m<sup>2</sup> in area, with 3 replications and 8 fertilizer treatments. Dry matter (DM) yields and mineral contents in soil and forage (P, K) were measured. The botanical composition was investigated each second year. Maximum DM yields were obtained by mineral NPK fertilization and a treatment called 'alternating fertilizer', with yearly alternating use of farmyard manure, liquid manure and mineral NPK. The application of farmyard manure compost reduced DM yields. The additional application of stone-meal and metallurgical lime to slurry did not increase the effects of untreated slurry on yield. Fertilization with slurry increased the proportions of grasses, whereas farmyard manure increased forbs. The proportion of legumes was increased by PK and by fertilization with slurry with lime.

Keywords: slurry, stable manure, compost, organic farming, mowing-pasture

## Introduction

Environmentally friendly use of organic fertilizers requires that nutrients are used with high efficiency. In particular, applications of slurry need to be exact both in their timing and amount. This has been well known for a long time, but is not always realised in practice. Objections against long-term application of slurry exist with regard to negative effects on soil microflora or because of leaf burn of grassland plants (Normann-Schmidt, 1993), which can influence the floristic composition of grassland swards. Addition of stone meal or metallurgical lime, as well as fertilization with farmyard manure or farmyard manure compost seems to be easier to handle particularly in land use systems using the biodynamic management. The objective of this experiment was therefore the investigation of long term effects of different systems of conventional and organic fertilization on soil minerals, dry matter yields and botanical composition, comparing both cut and grazed grassland.

## Materials and methods

The experiment was carried out on a permanent grassland in Southwest Germany on an alluvial soil (mean yearly temperature 7.0 °C; mean precipitation 1,000 mm). The experimental design was a split plot design with 8 fertilizer treatments (Table 1) in 3 replications under cutting as well as under mowing pasture use; size of subplots was 25 m<sup>2</sup>. Botanical composition was assessed every two years. Dry matter yield percentages of all species were estimated in the second growth. Yield data were subjected to an ANOVA by the procedure 'mixed' of the Software SAS/Stat 9.1 and means were compared using the LSD-test. Soil mineral contents (0-10cm soil depth) were measured in each autumn with the CAL method (VDLUFA, 2000). The determination of N mineralization in the topsoil (only in 2004) follows the anaerobic incubation after Kandeler (1993). The results gave an overview of the actual activity of the soil microflora for N-mineralization.

Table 1. Fertilizer treatments and mean nutrient input in kg ha<sup>-1</sup> y<sup>-1</sup> (Subsequent delivery of nutrients from excrements under grazing was not taken into account).

Number and Treatment	Mean total nutrient input N/P/K (kg ha <sup>-1</sup> y <sup>-1</sup> )
1 mineral fertilizer	160/52/166
2 mineral fertilizer	0/52/166
3 farmyard manure (2 x 16 t ha <sup>-1</sup> y <sup>-1</sup> ) in yearly alternation with liquid manure (2 x 40 m <sup>3</sup> y <sup>-1</sup> )	109/23/137
4 farmyard manure compost (2 x 16 t ha <sup>-1</sup> y <sup>-1</sup> )	159/43/168
5 Yearly alternating fertilization – farmyard manure – liquid manure – mineral fertilizer	126/33/139
6 slurry (3 x 30 m <sup>3</sup> to 2 <sup>nd</sup> and 4 <sup>th</sup> and after 4 <sup>th</sup> regrowth)	172/27/216
7 slurry (3x analogue to treatment 6) and additional stone meal (60 kg ha <sup>-1</sup> ) to 1 <sup>st</sup> and 3 <sup>rd</sup> regrowth	172/27/216
8 slurry (3x analogue to treatment 6) and additional metallurgic lime (60 kg ha <sup>-1</sup> ) to 1 <sup>st</sup> and 3 <sup>rd</sup> regrowth	172/27/216

## Results and discussion

The changes of phosphorus and potassium contents of the topsoil varied between the treatments, but there were no big differences in P between grazed and cut treatments (Table 2). Even though the mineral fertilization treatments received the highest P inputs this did not result in higher P contents in the topsoil after 20 years. K contents decreased in the mineral fertilizer treatments and increased markedly for treatment 3, under grazing and under cutting.

Table 2. Development of plant available P and K contents in the soil (after CAL method) during the experimental period (from 1985 to 2004) in mg kg<sup>-1</sup>.

Treatment	P (mg kg <sup>-1</sup> )	P (mg kg <sup>-1</sup> )	P (mg kg <sup>-1</sup> )	P (mg kg <sup>-1</sup> )	K (mg kg <sup>-1</sup> )	K (mg kg <sup>-1</sup> )	K (mg kg <sup>-1</sup> )	K (mg kg <sup>-1</sup> )
Management	meadow	meadow	pasture	pasture	meadow	meadow	pasture	pasture
year	1985	2004	1985	2004	1985	2004	1985	2004
1	1.7	3.0	2.6	4.2	10.0	7.9	19.1	12.7
2	2.2	4.1	2.2	4.5	13.3	12.2	16.6	13.5
3	4.4	1.5	5.2	1.9	10.0	23.5	15.8	27.4
4	3.9	4.8	3.9	5.4	12.5	9.0	16.6	12.5
5	4.8	2.1	3.1	2.7	11.6	14.1	14.1	16.0
6	3.5	2.1	3.1	2.8	12.5	10.5	14.9	14.9
7	3.5	2.3	3.5	2.8	12.5	13.3	19.1	15.8
8	2.6	3.4	2.2	3.5	14.9	19.7	18.3	18.0

The potential nitrogen mineralization is shown in Table 3. Obviously, the treatments with mineral nitrogen fertilization (treatment 1 and 5) have only low nitrogen mineralization.

Table 3. Potential for nitrogen mineralization measured in soil depth 0-10 (µg N g DM<sup>-1</sup> d<sup>-1</sup>) (we gratefully thank Dr. H. Flaig, LTZ Augustenberg for the investigation of this parameter).

Treatment	1	2	3	4	5	6	7	8
Meadow	5.46	8.93	8.79	8.08	6.29	6.88	8.22	7.35
Mowing	7.96	11.03	11.95	11.74	9.80	11.01	10.94	11.01
pasture								

Treatment 3 (alternating fertilization) had a higher nitrogen mineralization potential both under mowing and grazing, while for treatment 2 (PK fertilization) N mineralization was only significantly higher under cutting, and for treatment 4 (farmyard manure compost) only under

grazing. Generally the mineralization capacity was higher for grazed plots, whereas the relationships between the eight treatments were very similar.

Comparing the first and last experimental year, great differences in the percentage of grasses and legumes could be observed (Table 4). Highest increase of grasses resulted by mineral nitrogen fertilization. Legumes were mainly promoted by 'PK' and 'slurry-with-lime'.

Table 4. Percentage of dry matter yield of grasses, legumes and forbs comparing the years 1985 and 2004. Different letters denote statistically different means between treatments; asterisks mark the significance between the mean percentages in the two years ( $P = 0.05$ ).

Treatment	Grasses			Legumes			Forbs		
	1985	2004		1985	2004		1985	2004	
1	37.8 a	73.7 a	***	8.7 a	2.1 d		53.5 a	24.3 a	***
2	31.7 a	41.3 c		15.5 a	28.0 a	**	52.8 a	30.7 a	***
3	39.2 a	54.2 b	**	12.5 a	11.7 cd		48.3 a	34.2 a	*
4	33.8 a	50.3 bc	***	14.5 a	13.7 c		51.7 a	36.0 a	**
5	33.8 a	59.7 b	***	14.8 a	9.7 cd		51.2 a	30.7 a	***
6	33.0 a	53.0 bc	***	14.3 a	15.0 bc		52.7 a	32.0 a	***
7	32.7 a	55.3 b	***	13.0 a	12.5 cd		54.3 a	32.2 a	***
8	33.5 a	51.3 bc	***	12.3 a	25.3 ab	**	54.2 a	23.3 a	***

Highest dry matter yields were observed generally for the mowing pasture, which may be caused by the additional nutrient input via excrements. Highest dry matter yields were observed by mineral fertilizer (treatment 1) and slurry (treatment 6), whereas lowest yields occurred for PK (treatment 2). The addition of stone meal or lime to slurry had no positive effects on yields.

Table 5. Harvested biomass yields ( $\text{t DM ha}^{-1}\text{y}^{-1}$ ) for the eight treatments (1984-2004). Different letters denote statistically different means within the management system ( $P = 0.05$ ).

Treatment	1	2	3	4	5	6	7	8
meadow	10.20 a	8.77 c	9.54 b	9.28 b	10.11 a	10.13 a	9.49 b	9.42 b
mowing pasture	12.03 a	10.72 d	11.34 bc	11.52 b	11.92 a	11.08 cd	11.18 bc	11.18 bc

## Conclusions

Comparisons between different fertilizing systems are particularly valuable, especially if they cover a long period as in the case of this experiment. Organic fertilizing (treatments 3, 4, 6-8) or the abandonment of mineral nitrogen resulted in lower herbage DM yields compared with mineral NPK treatment or the treatment with alternating fertilizer. The percentage of grasses was increased by fertilizing with mineral nitrogen, and that of legumes by fertilization with PK and slurry-with-lime.

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# Apparent balance of nitrogen in organic and conventional dairy farms in Tuscany (Central Italy)

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## Abstract

Data obtained from a survey conducted on 36 organic and conventional dairy farms in Mugello (Tuscany, Central Italy) are reported. The apparent balance of nitrogen (N) was calculated for each farm in order to evaluate the efficient utilization of this element, comparing all inputs and all outputs of different forms of N entering or leaving the farm. The surplus of the element was then related to the farm surface area to compare farms of different size. Organic farms are located at a higher altitude and have a larger land surface than conventional ones. Large differences were found concerning land use, with a greater use of maize (16.1% vs. 2.4%,  $P < 0.05$ ) and a reduced presence of natural pastures (5.9% vs. 15.7%,  $P < 0.05$ ) in the conventional farms. The apparent lower surplus of N on organic farms than conventional farms was not significant, and this may be related to purchased fertilizers ( $P < 0.05$ ). The efficiency of N utilization was similar in organic (27.3%) and conventional farms (22.5%). Results show that stocking rate was the main factor that affected N surplus and there were no significant differences in N surplus between organic and conventional farms. It can be concluded that conventional dairy farms in Mugello are managed in an extensive manner with no high environmental risks, particularly concerning N pollution.

Keywords: apparent nitrogen balance, dairy farms, organic farming, stocking rate

## Introduction

The evaluation of sustainability of an animal production system can be performed through an estimation of the risks of nitrogen (N) losses, which can be assessed by the apparent balance method (Simon and Le Corre, 1992). According to this methodology, the fluxes of the different forms of the nutrient entering or leaving the farm are calculated in order to compute the surplus and this is expressed on a surface-area basis to compare farms of different size. It is then possible to evaluate possible technical improvements to reduce waste and risks of pollution. At the same time it is necessary to maintain efficient animal husbandry, taking into account both the exploitation of resources and the need to supply the animals' nutritive requirements, in order to obtain effective and productive outcomes. The aim of this work is to compare the organization of production in conventional and organic dairy farms in an area of Tuscany (Central Italy) in order to evaluate the apparent balance of nitrogen in two animal husbandry systems.

## Materials and methods

The survey was conducted in the area of Mugello (North Tuscany) on 36 dairy farms representative of the conventional and organic farming systems (9 and 27 farms, respectively) present in the district. For each farm, data were collected concerning land use, size of the flock and all the forms of nitrogen (N) entering or leaving the farm, such as animals, feedstuffs, fertilizers and different kind of vegetal material (e.g. hay or straw). N-fixation was estimated as 100 kg N ha<sup>-1</sup> for pure stands of legumes (mainly lucerne) and as 15 kg N ha<sup>-1</sup> for mixtures and natural pastures, according to a previous study carried out in the same area (Argenti *et al.*,

1996). The amounts of N have been turned into fluxes of the element to calculate the apparent balance utilizing, where possible, direct transformation coefficients (e.g. analysis of milk and forage crops, feedstuffs contents) or using estimations or tables (Borgioli, 1981; Tamminga, 1992; Van Horn *et al.*, 1994). Analysis of the data to compare the two farm typologies was performed using the GLM procedure of SAS software (1988), following this model:

$Y_{ij} = \mu + B_i + E_{ij}$ , where the fixed factor is the farm typology.

## Results and discussion

Organic farms are at slightly higher elevation than conventional ones and they present a greater land surface area (Table 1). Land use is remarkably different between typologies, particularly the greater use of maize on conventional farms (16.1% vs. 2.4%) whereas organic farms have a greater use of forage crops, especially natural pastures (15.7% vs. 5.9%).

Table 1. Main characteristics of studied farms (average  $\pm$  standard error).

	Organic farms	Conventional farms	Significance
Average height (m a.s.l.)	586 $\pm$ 58.46	447 $\pm$ 33.75	*
Total surface (ha)	127 $\pm$ 19.61	50 $\pm$ 11.32	**
Agricultural area (ha)	95 $\pm$ 12.19	39 $\pm$ 7.04	**
Land use (%)			
Winter cereals	12.6 $\pm$ 3.41	12.6 $\pm$ 1.97	ns
Maize	2.4 $\pm$ 5.69	16.1 $\pm$ 3.28	*
Other crops	1.9 $\pm$ 1.50	1.4 $\pm$ 0.87	ns
Total arable crops	16.9 $\pm$ 8.02	30.1 $\pm$ 4.63	ns
Lucerne	14.7 $\pm$ 6.33	21.6 $\pm$ 3.65	ns
Meadows	26.8 $\pm$ 10.35	24.0 $\pm$ 5.97	ns
Artificial pastures	25.9 $\pm$ 8.99	18.4 $\pm$ 5.19	ns
Natural pastures	15.7 $\pm$ 4.33	5.9 $\pm$ 2.50	*
Total forage crops	83.1 $\pm$ 8.02	69.9 $\pm$ 4.63	ns
Stocking rate (LU ha <sup>-1</sup> year <sup>-1</sup> )	0.87 $\pm$ 0.13	0.95 $\pm$ 0.65	ns
Number of cows	55.9 $\pm$ 8.76	24.9 $\pm$ 5.06	**
Days of grazing	129 $\pm$ 27.46	96 $\pm$ 15.85	ns
Farm milk production (kg year <sup>-1</sup> )	361411 $\pm$ 82160	137843 $\pm$ 47435	*
Cow milk production (kg year <sup>-1</sup> )	5186 $\pm$ 790	5228 $\pm$ 456	ns
Milk surface production (kg ha <sup>-1</sup> )	4315 $\pm$ 1238	3844 $\pm$ 715	ns

ns = not significant; \* =  $P < 0.05$ ; \*\* =  $P < 0.01$ .

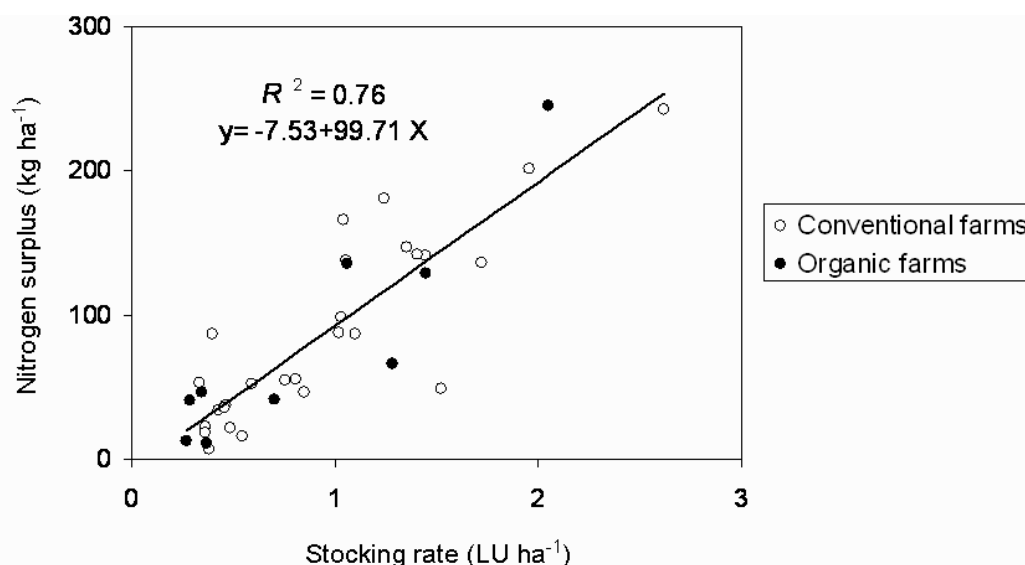


Figure 1. Relationship between stocking rate and nitrogen surplus.



Moreover, organic farms have a greater number of cows even though the stocking rate is rather low in both systems and not significantly different between them. Organic farms are characterized by a higher milk production, due mainly to the greater number of cows; the average cow production is not significantly different from conventional ones.

Total inputs of N are rather low in both typologies and values of surplus of the element are not significantly different between organic and conventional farms (Table 2). The factor that mainly affects N surplus is the purchase of fertilizers, which is absent in the organic farms. Efficiency in the use of N appears to be slightly higher in organic farms, though the difference is not significant. Level of stocking rate is strongly related to surplus of N and the regression (Figure 1) presents a high coefficient of determination utilizing data from both farm typologies.

Table 2. Nitrogen apparent balance ( $\text{kg ha}^{-1}$ ) for the two farm typologies.

	Organic farms	Conventional farms	Significance
<b>Inputs (+)</b>			
Animals	0.4	0.8	ns
Fertilizers	0.0	28.9	*
Feedstuffs	87.7	54.3	ns
N-fixation	22.8	28.2	ns
<i>Total inputs</i>	110.9	112.2	ns
<b>Outputs (-)</b>			
Vegetal products	3.5	2.6	ns
Animals	2.6	3.0	ns
Fertilizers	1.1	0.3	ns
Milk	23.1	19.3	ns
<i>Total outputs</i>	30.3	25.2	ns
Surplus (Inputs-Outputs)	80.6	87.0	ns
Efficiency (%)	27.3	22.5	ns

ns = not significant; \* =  $P < 0.05$ ; \*\* =  $P < 0.01$ .

## Conclusions

The survey permitted the characterization of the two farm typologies and the identification of the main differences in land use between the different production systems. No difference in N surplus was observed, so it is possible to state that the conventional farms in the studied area are also managed in an extensive way, and the environmental risks are very limited compared with other animal husbandry systems that are characterized by higher stocking rates.

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## Effect of bio and mineral fertilization on fresh weight and dry matter forage yields of three pearl millet varieties

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### Abstract

Experiments were carried out at the Forage Crops Research Department, Giza Research Station, during the summer of 2002 and 2003, to evaluate the response of three varieties of pearl millet (*Pennisetum americanum*) (Shandaweel-1, Synthetic and Niger) to different levels of nitrogen fertilization (0, 71.4, 142.8 and 285.6 kg N ha<sup>-1</sup>) alone and in combination with composite inocula. Inocula contained nitrogen-fixing bacteria (*Klebsiella pneumoniae*, *Pseudomonas putida*, *Azospirillum lipoferum*, *Azotobacter chroococcum* and *Bacillus megatherium*). Over the two years responses to various applications, in terms of fresh weight and dry matter (DM) yields, were significantly greater than yields from the control (102.1 t ha<sup>-1</sup> for fresh yield and 17.3 t ha<sup>-1</sup> for DM yield). The highest response was obtained from the application of 285.6 kg N ha<sup>-1</sup> (variety mean was 145.6 t ha<sup>-1</sup> for fresh yield, and 25.0 t ha<sup>-1</sup> for DM yield). The application of 142.8 kg N ha<sup>-1</sup> in combination with composite inocula ranked second, with 136.7 t ha<sup>-1</sup> for fresh yield and 24.0 t ha<sup>-1</sup> for DM yield. Average treatment yields for Shandaweel-1 exceeded the treatment means of the other two varieties in fresh yield (130.4 t ha<sup>-1</sup>) and in DM yield (22.0 t ha<sup>-1</sup>).

Keywords: pearl millet, biofertilization, dry matter yield

### Introduction

In Egypt, the production of green forage is less than the demand to support meat and milk production. Pearl millet (*Pennisetum americanum*) is considered one of the most important summer multi-cut forage crops. The cultivated area has stimulated yearly increases to meet the requirements for animal feed. Large amounts of nitrogen fertilizers have been used to produce high yields of forage millet. Attention has also been given to pollution reduction, and great efforts have been exerted in order to produce products without nitrate pollution, while also maintaining a high yield potential. Biofertilization is an approach to organic cultivation which has provided significant improvements in growth, dry matter and protein yield of forage grasses. It was achieved by inoculation with *Azospirillum* in field and greenhouse experiments (Okon *et al.*, 1983; Yahalon *et al.*, 1984). Phosphate-solubilizing microorganisms (PSMs), which are ubiquitous in soils, could play an important role in supplying P to the plants in a more friendly and sustainable manner (Gyaneshwar *et al.*, 2002).

The aim of this investigation was to study the effect of inorganic fertilizer (N), and inoculation with composite inocula of nitrogen fixers and phosphate-dissolving bacteria under different levels of nitrogen fertilization, and the yield response to such treatments for three different varieties of pearl millet.

### Materials and methods

Two experiments were conducted at the Agricultural Research Centre, Giza, Egypt during the summer of 2002 and 2003 to study the yield response of three different varieties of pearl millet to inoculation with composite inocula of biofertilizers under different levels of mineral

nitrogen (0.0, 71.4 and 142.8 kg N ha<sup>-1</sup>) compared with four N treatments without inoculation (0.0, 71.4, 142.8 and 285.6 kg N<sup>-1</sup>).

The experimental design was a split-plot with four replicates. The three pearl millet varieties (two local varieties (Shandaweel-1 and Giza synthetic)) and an imported variety (Niger) were assigned to the main plots and the fertilization treatments were arranged in the subplots. The subplot size was 12 m<sup>2</sup> (3 x 4 m). Seeds for inoculated plots were soaked for one hour before planting in a mixed broth culture of strains containing 10<sup>9</sup> cells ml<sup>-1</sup>. One week later, spray application of the liquid inocula was added along a groove made in close vicinity to the row of germinated seedlings. The composite of bacterial strains consisted of five strains, *Klebsiella pneumoniae*, *Pseudomonas putida*, *Azospirillum lipoferum*, *Azotobacter chroococcum* and *Bacillus megatherium*, which were provided by the Microbiology Department, Faculty of Agriculture, Cairo University, Egypt. The seeds were hand drilled in rows 20 cm apart at the seeding rate of 35.7 kg ha<sup>-1</sup>. Nitrogen fertilizer in the form of ammonium sulphate (20.6% N) was divided into three equal doses. Three cuts were obtained in each season at 50, 85 and 120 days after planting. Totals of fresh and dry matter yield (t ha<sup>-1</sup>) for three cuts were recorded. Statistical analysis was carried out for fresh and dry matter yield of individual cuts, as well as total seasonal yield, according to Steel and Torrie (1980).

## Results and discussion

The combined analysis over the two seasons is presented in Table 1. The data demonstrate the differences between the three studied varieties as affected by different levels of nitrogen, biofertilization, and their mixtures. Significant differences were found between the tested varieties as well as between treatments, in terms of fresh weight and dry matter forage yields. Significantly greater production was found for the variety Shandaweel-1 than the synthetic and Niger varieties, in fresh yield (130.4, 124.8 and 118.1 t ha<sup>-1</sup>) and dry matter yield (22.0, 20.9 and 19.9 t ha<sup>-1</sup>), respectively. These results reveal that Shandaweel-1 surpassed the synthetic and Niger varieties by 5.6 and 12.3 t ha<sup>-1</sup> for fresh yield, and 1.1 and 2.1 t ha<sup>-1</sup> for dry matter yield over two growing seasons.

Table 1. Total fresh and dry matter forage yields (t ha<sup>-1</sup>) of three pearl millet varieties under different levels of nitrogen and biofertilization over two seasons.

Varieties	Fresh forage yield				Dry matter forage yield			
	Shand.	Synth.	Niger	Mean	Shand.	Synth.	Niger	Mean
Treatments								
Without N	101.8 <sup>f</sup>	105.2 <sup>de</sup>	99.3 <sup>c</sup>	102.1 <sup>f</sup>	16.5 <sup>c</sup>	17.6 <sup>c</sup>	17.7 <sup>d</sup>	17.3 <sup>e</sup>
Inoculation	113.5 <sup>g</sup>	107.8 <sup>d</sup>	103.3 <sup>c</sup>	108.1 <sup>e</sup>	18.3 <sup>b</sup>	19.2 <sup>b</sup>	17.9 <sup>c</sup>	18.5 <sup>d</sup>
71.4 kg N ha <sup>-1</sup>	121.7 <sup>e</sup>	121.8 <sup>c</sup>	119.8 <sup>b</sup>	121.1 <sup>d</sup>	20.1 <sup>b</sup>	20.4 <sup>b</sup>	17.5 <sup>c</sup>	19.4 <sup>d</sup>
142.8 kg N ha <sup>-1</sup>	138.6 <sup>cd</sup>	130.2 <sup>b</sup>	128.7 <sup>a</sup>	132.5 <sup>c</sup>	24.0 <sup>b</sup>	20.2 <sup>b</sup>	19.9 <sup>b</sup>	21.4 <sup>c</sup>
285.6 kg N ha <sup>-1</sup>	150.3 <sup>a</sup>	145.7 <sup>a</sup>	119.5 <sup>b</sup>	145.6 <sup>a</sup>	24.9 <sup>a</sup>	24.5 <sup>a</sup>	25.6 <sup>a</sup>	25.0 <sup>a</sup>
Inoc. + N1	142.1 <sup>bc</sup>	130.8 <sup>b</sup>	123.5 <sup>b</sup>	132.4 <sup>c</sup>	24.4 <sup>b</sup>	20.5 <sup>b</sup>	18.4 <sup>b</sup>	21.1 <sup>c</sup>
Inoc. + N2	144.8 <sup>b</sup>	132.1 <sup>b</sup>	133.1 <sup>a</sup>	136.7 <sup>b</sup>	25.8 <sup>a</sup>	23.8 <sup>a</sup>	22.3 <sup>a</sup>	24.0 <sup>b</sup>
Average	130.4	124.8	118.1	125.5	22.0	20.9	19.9	21.0
LSD 0.05		1.9				0.7		

N1 = 71.4 kg N ha<sup>-1</sup> and N2 = 142.8 kg N ha<sup>-1</sup>.

Treatments using mineral N-fertilization and bacterial inoculation showed significant effects on fresh and dry matter yields. The treatment with 285.6 kg N ha<sup>-1</sup> produced the highest fresh (145.6 t ha<sup>-1</sup>) and dry matter (25.0 t ha<sup>-1</sup>) forage yields over two seasons.

The application of 142.8 kg N ha<sup>-1</sup> combined with the composite inocula of associated diazotrophs and phosphate-dissolving bacteria resulted in higher dry matter forage yield (24.0 t ha<sup>-1</sup>) compared with the 71.4 and 142.8 kg N ha<sup>-1</sup> treatments without biofertilization. The variety Shandaweel-1 showed significant positive response to inoculation with

diazotrophs in the presence of 71.4 kg N ha<sup>-1</sup> for fresh weight (142.1 t ha<sup>-1</sup>) and dry matter (24.4 t ha<sup>-1</sup>) forage yield compared with the same treatment without inoculation (121.7 and 20.1 t ha<sup>-1</sup> for fresh and dry matter yields, respectively). This treatment (71.4 kg N ha<sup>-1</sup> + Inoc.) produced fresh and dry matter yields comparable to the treatment with 142.8 kg N ha<sup>-1</sup>. An increase in forage yield with associated diazotrophs and P-dissolving bacteria can be attributed not only to their N<sub>2</sub>-fixing proficiency but also to their ability to produce plant growth hormones (Dobereiner, 1977; Desale *et al.*, 1999).

It is well known that reductions in the use of nitrogen fertilizer are economically advantageous, with potential cost savings and energy reductions from using biological N-fixation. Biological fertilization assists the government and universities in increasing production with economic prices, in order for the farmer to reduce his expenditure and to reduce the use of nitrogen fertilizers, and is an important means for lowering nitrate pollution and keeping the environment clean.

## Conclusions

The application of a biofertilization culture to a crop of pearl millet (variety, Shandaweel-1), combined with a medium-rate application of N-fertilizer, i.e. 71.4 or 142.8 kg N ha<sup>-1</sup>, resulted in improved production compared with using fertilizer-N alone. Biofertilization seems to be beneficial in terms of enabling reduced fertilizer-N application rates and may have potential for pollution reduction as well.

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# The application of image analysis to estimate legume contents in legume/grass swards

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## Abstract

Under field conditions the botanical composition of mixed swards shows tremendous variation, affecting several important agronomic parameters (e.g. yield, forage quality). The present paper demonstrates perspectives and limitations of image analysis to assess the botanical composition of legume / grass mixtures without disturbing the growth of swards. Based on data from a pot experiment with binary mixtures of different seeding rates of red clover, white clover, lucerne and ryegrass, image analysis provided a promising tool for the estimation of legume contribution in swards. Using greyscale pictures of the swards, legume coverage predicted by image analysis showed a correlation with the measured DM contribution of legumes. Difficulties with visible bare soil in swards with a low tiller density could be avoided by integrating colour information into the image analysis procedure.

Keywords: digital image analysis, threshold, legume / grass swards, legume content

## Introduction

The composition of legume/grass swards affects numerous and important performance parameters like nitrogen fixation and yield. In this context the fraction of legumes is of high importance due to their ability to fix nitrogen. Using image analysis an efficient, indirect and non-destructive method should be developed to determine the botanical composition of swards, particularly the proportion of legumes. An image analysis procedure to estimate the cover percentage of legumes in images of the swards was generated with the commercial software Optimas<sup>®</sup> (Himstedt *et al.*, 2006). Misclassifications can be caused by weeds with rounded leaves (e.g. *Stellaria media*) and visible ground (Himstedt *et al.*, 2007). To improve the accuracy of the image analysis procedure the HSL (hue, saturation, lightness) colour model could be used to detect bare soil.

## Materials and methods

In 2004, a nine-week duration pot experiment with pure swards and binary legume/grass mixtures of red clover (*Trifolium pratense*), white clover (*Trifolium repens*), lucerne (*Medicago sativa*) and perennial ryegrass (*Lolium perenne*) was conducted. Eight experimental swards were investigated: pure swards of ryegrass (G: 20 kg ha<sup>-1</sup>), red clover (R: 8 kg ha<sup>-1</sup>), white clover (W: 4 kg ha<sup>-1</sup>) and lucerne (L: 16 kg ha<sup>-1</sup>) and the four binary mixtures red clover/ryegrass (R8 G8/20 kg ha<sup>-1</sup> and R2 G2/20 kg ha<sup>-1</sup>), white clover/ryegrass (WG 4/20 kg ha<sup>-1</sup>) and lucerne/ryegrass (LG 16/20 kg ha<sup>-1</sup>). To compare swards of different ages the legume/grass mixtures were harvested 21, 35, 49 and 63 days after sowing. All treatments were sown in four replicates (n = 64). The sowing was done manually in a distance between rows of 12 cm and at a sowing depth of 0.5 cm. The sward size was 0.119 m<sup>2</sup>. The pots were filled with 2 cm drainage substratum (Lavagrus) and about 16 cm homogenized loamy soil (sL- 1S; 3.6%, 73% U, 23.4% T and 2% humus). No fertilizers were used. To determine the sward composition, total aboveground biomass was fractionated in grass, legumes and unsown species. The samples were dried for 24 hours at 65° C. One day before the harvest

digital pictures of the swards were taken with a Canon Power Shot G6 Digital Camera. Flashlight was used for all pictures to ensure uniform illumination. For image analysis the image processing software Optimas<sup>®</sup> was used (Media Cybernetics, 1999). The image analysis procedure was arranged as follows. (1) Import of the images in tiff format with 766 x 744 pixels. (2) Conversion of the digital colour images into 8-bit greyscale images. (3) Run the optimal image analysis procedure with the best erode/dilate combination and a greyscale threshold range from 71.0 to 145.0. (4) The result is the legume covered area ascertained by image analysis  $P_o$  (%). (5) The assumption is that with this measured cover percentage of legumes it is possible to suggest the dry matter (DM) proportion of legumes. To identify misclassifications the identified legume area was made visible as layer and fused with the original colour image. Misclassifications of the three categories unsown species, grass and bare soil were detected within the area, which was estimated as legume covered area. To identify visible bare soil by image analysis, 24-bit HSL colour images (hue, saturation, lightness) of the swards were ascertained. For image segmentation into bare soil and plants, a histogram segmentation with thresholding was used. As reference the actual area of bare soil in each image was estimated. To measure the actual area of bare soil in each image the optimal threshold to distinguish soil and plants was ascertained respectively. The actual area of bare soil ( $BS_a$  %) and the threshold for H, S and L was noted. The average of all thresholds was computed and used as basis for the automatic image analysis procedure to measure the bare soil area.

## Results and discussion

With the image analysis it is possible to measure the cover percentage of legumes. A relationship between cover percentage and DM contribution exists ( $R^2 = 0.65$ , s.e. 12.0) including all legume species of the 49 and 63 days old swards (Figure 1).

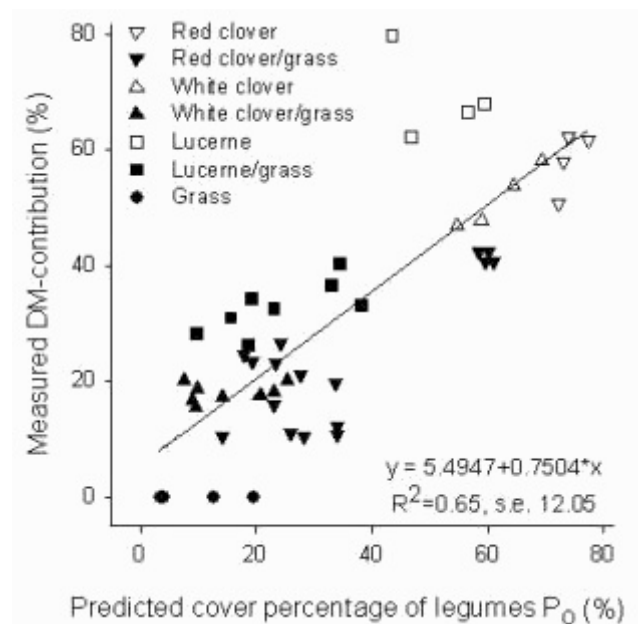


Figure 1. Coherence between the cover percentage and the DM-contribution of legumes (49 and 63 days old swards).

It is remarkable that red clover and white clover pure swards and mixtures show a closer relationship with the reference than lucerne and grass. Separate correlations for individual legume species produce higher accuracies with  $R^2 = 0.97$  (s.e. 4.0) for white clover,  $R^2 = 0.83$

(s.e. 7.6) for red clover and  $R^2 = 0.71$  (s.e. 10.5) for lucerne. The evaluation of the sources of error shows that visible bare soil leads to important misclassifications. In younger and more open swards the misclassification of bare soil as legume is quite high. On average, 24 per cent of the area classified as legume is visible bare soil, with a range from 0.29 to 91.59 per cent. A misclassification of grass (on average 15.1 per cent of the area classified as legumes) and unsown species (on average 6.6 per cent) can be found in swards of all ages. The chance to misclassify grass as legume is greater than of misclassifying unsown species as legume, because grass is present in all mixtures. To detect the bare soil area HSL colour images were used. The average value of the 64 image-specific colour thresholds range for hue from 18 to 45, for saturation from 43 to 115 and for lightness from 70 to 173. Applying these thresholds on all 64 images improved the accuracy of discrimination bare soil ( $BS_p$ ) and legume cover. Bare soil areas between 0.3 per cent to 81.4 per cent were detected and the correlation between  $BS_a$  and  $BS_p$  ( $R^2 = 0.99$ ; s.e. 2.82) allows an accurate prediction of the percentage of bare soil areas in sward images (Figure 2).

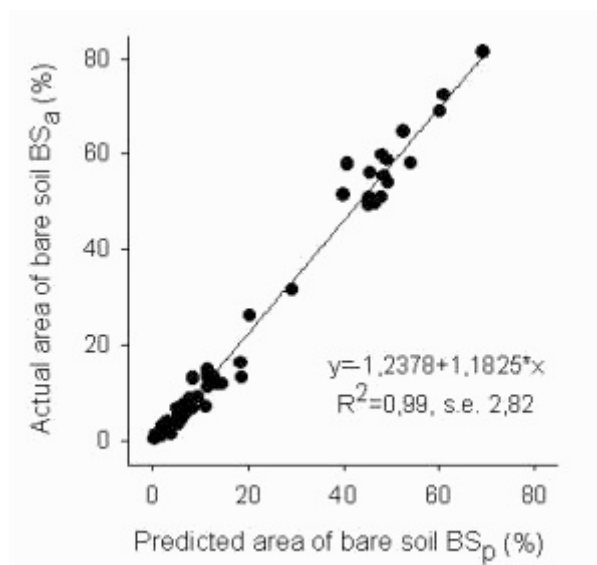


Figure 2. Correlation between the actual ( $BS_a$ ) and the predicted ( $BS_p$ ) percentage of bare soil areas in sward images.

## Conclusions

Using image analysis to measure the legume covered area in sward images and the DM contribution of legumes seems to be very promising. The outcomes suggest that a legume-specific approach produces better results. To improve the accuracy of the estimation of legume DM contribution, the integration of colour information is important. The HSL colour model is useful particularly for the discrimination of bare soil and plant covered area.

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# Balancing milk yield and stocking rate to the output from an organic crop rotation system

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## Abstract

An organic crop rotation system designed for supplying all feed to a herd of dairy cattle was run during 1993-2006 in Central Norway. During the second course of rotation (2003-2006) the yields of forage (two years with ley) and grain crops (barley and oats with peas) from the system were estimated sufficient to feed a herd with all energy needed for a production level of up to 8000 kg milk cow<sup>-1</sup> year<sup>-1</sup> given a stocking rate of 0.7 ha<sup>-1</sup>. However, due to low contents of crude protein in the harvested plants, a production level of 5,000-6,000 kg ha<sup>-1</sup> yr<sup>-1</sup> seemed more feasible, especially if leys are cut late and/or infrequently.

Keywords: energy supply, feed, organic milk production, protein supply, self sufficiency

## Introduction

An organic crop rotation system designed for supplying all feed to a herd of dairy cattle was established in 1993 (Bakken *et al.*, 2006). During the first course of rotation (undersown spring barley, two cuts from 1st, 2nd and 3rd-year ley, swede and oats) ending in 1999, the yields of barley and the total protein supply was too low to sustain the target milk yield of 6,000 kg cow<sup>-1</sup> year<sup>-1</sup> at a stocking rate of 1.0 ha<sup>-1</sup>. This led to changes in the proportion and type of grain crops in the rotation and the management of leys. The aim was to see if further improvements and stabilization of the supply of protein and energy to the herd were possible, and to gain knowledge of what yield levels and stocking rates are feasible in the region where the system is located, which is one of the most important regions for organic milk production in Norway. In the present paper this topic is discussed on the basis of yields obtained in the system from 2003 to 2006.

## Materials and methods

The cropping system was located in Stjørdal, Norway (63°N, 8°E). Management and yields during the period 1993-1999 has been described by Bakken *et al.* (2006). Since 2003 the system has consisted of eight 1,400 m<sup>2</sup> plots for two replicates of a rotation of undersown spring barley, 1st and 2nd year ley and a bicrop of oats and peas. The sown species in the leys were *Phleum pratense*, *Festuca pratensis*, *Lolium perenne*, *Poa pratensis*, *Trifolium pratense*, *T. repens* and *T. hybridum*. All plots were further split into two subplots for two different harvesting regimes in the years with leys. The 'intensive' management involved a first cut taken at late stem elongation of *P. pratense*, a second cut 600 day° (heat sum units, base temperature 0°C) later and a third one in mid September. 'Extensive' management involved a first cut taken at heading of *P. pratense* and a second one in late August. Machinery for full scale farming was used for tillage, fertilization, sowing and harvesting. Samples for determination of yield quantity and quality were harvested from three randomly chosen 10 m<sup>2</sup> rectangles within each subplot. Half of the manure amounting 0.7 animal unit ha<sup>-1</sup> was supplied to barley, and the rest evenly distributed between the other crops. The data presented

for forage quality and content of clover are as analysed by NIRS (Lunnan and Marum, 1994; Fystro and Lunnan, 2006). Evaluations of the fodder supply were based on Norwegian standards (Anonymous, 1997) for net energy (milk feed units, FEm) and protein (amino acids absorbed in the intestine, AAT) requirements for adult cows (550 kg liveweight and one pregnancy year<sup>-1</sup>). In the computations, table values were used for the grains (www.mattilsynet.no). The loss of forage from field to animal intake was regarded as being 20% of recorded DM yields. The first year (2003) of the rotation was omitted from the analyses as all leys were sown that year and the yields from them were not regarded as representative of the potential.

## Results and discussion

Improved yields of barley in particular, were obtained in the new rotation (3290 kg ha<sup>-1</sup> year<sup>-1</sup>, Table 1) compared to the previous (1500 kg ha<sup>-1</sup> year<sup>-1</sup>, Bakken *et al.* 2006). Also the yields of forage were higher during the period 2003-2006 compared to the period 1994-1999. The crude protein contents in the forages were generally low, third cuts (intensive management) excepted. Thus, even in second cuts containing as much as 50-60% clover (Table 1), the protein balance in the rumen (PBV) was negative in most years. Due to this, and the negative PBV-values of barley (www.mattilsynet.no), less than one-third of the net energy produced was associated with a positive PBV. However, contents of crude protein, AAT and FEm in forages were higher under 'intensive' compared to 'extensive' management.

Table 1. Yields (kg DM ha<sup>-1</sup>) from an organic cropping system in four years. Means  $\pm$  standard deviations (N = 4).

Year	Leys, intensive			Leys, extensive		Barley	Oats with peas	Prop. of peas
	1 <sup>st</sup> cut	2 <sup>nd</sup> cut	3 <sup>rd</sup> cut	1 <sup>st</sup> cut	2 <sup>nd</sup> cut			
2003						3100 $\pm$ 99	4550 $\pm$ 141	0.68
2004	4140 $\pm$ 710	3310 $\pm$ 690	1600 $\pm$ 280	5800 $\pm$ 850	5610 $\pm$ 1280	3920 $\pm$ 401	4150 $\pm$ 636	0.73
2005	2460 $\pm$ 260	2930 $\pm$ 500	2470 $\pm$ 570	5600 $\pm$ 830	3440 $\pm$ 560	2920 $\pm$ 91	2720 $\pm$ 489	0.63
2006	2460 $\pm$ 530	1860 $\pm$ 590	2810 $\pm$ 360	3670 $\pm$ 1040	3430 $\pm$ 340	2320 $\pm$ 422	4390 $\pm$ 392	0.56

Table 2. Nutritive value and clover content (kg DM<sup>-1</sup>) of forage produced under intensive (Int) or extensive (Ext) management. Means  $\pm$  standard deviations (N = 12; four each year during the period 2004-2006<sup>1</sup>).

	CP, g	NDF, g	INDF, g	DMD, g	FEm	AAT, g	PBV, g	Clover, g
Int, 1 <sup>st</sup> cut	122 $\pm$ 21	427 $\pm$ 47	49 $\pm$ 21	810 $\pm$ 34	0.99 $\pm$ 0.05	87 $\pm$ 34	-28 $\pm$ 18	240 $\pm$ 120
Int, 2 <sup>nd</sup> cut	129 $\pm$ 12	454 $\pm$ 29	92 $\pm$ 15	726 $\pm$ 25	0.85 $\pm$ 0.04	79 $\pm$ 23	-7 $\pm$ 11	470 $\pm$ 90
Int, 3 <sup>rd</sup> cut	176 $\pm$ 23	425 $\pm$ 59	92 $\pm$ 13	693 $\pm$ 13	0.83 $\pm$ 0.03	78 $\pm$ 20	42 $\pm$ 20	610 $\pm$ 200
Ext, 1 <sup>st</sup> cut	100 $\pm$ 19	527 $\pm$ 45	77 $\pm$ 6	736 $\pm$ 17	0.87 $\pm$ 0.03	80 $\pm$ 19	-37 $\pm$ 18	230 $\pm$ 160
Ext, 2 <sup>nd</sup> cut	118 $\pm$ 16	481 $\pm$ 31	129 $\pm$ 30	654 $\pm$ 27	0.76 $\pm$ 0.03	72 $\pm$ 21	-7 $\pm$ 16	600 $\pm$ 170

<sup>1</sup>Abbreviations: DM = dry matter, CP = crude protein, NDF = neutral detergent fibre, INDF = indigestible NDF, DMD = dry matter digestibility, FEm = milk feed units, AAT = amino acids absorbed in the intestine, PBV = protein balance in the rumen.

The forage and grain crops supplied from the system in 2004-2006 were estimated as sufficient to feed a herd with all FEm and AAT needed for a production level up to 8000 kg milk cow<sup>-1</sup> year<sup>-1</sup> given a stocking rate of 0.7 ha<sup>-1</sup> (Figure 1). One presumption is then a constant milk yield throughout the lactation period (26 kg day<sup>-1</sup>). Usually, milk yields are higher early in the lactation than late, and consequently also the nutrient requirements. Although apparently sufficient supply of FEm and AAT even at 35 kg milk day<sup>-1</sup>, the PBV of the total ration was negative and strongly so for rations based on extensively managed leys (< - 500 g day<sup>-1</sup>, results not shown). According to Norwegian standards, PBV should be zero

at peak lactation. This indicates that intensive management is favourable compared to extensive management aiming high milk yields. However, low concentrations of phosphorous and sulphur in the forage may cause insufficient supply of these elements for high yielding cows (results not shown). Furthermore, even though peas contributed significantly to a higher AAT supply in this rotation compared to the previous, the frequency of peas was too high regarding the risk of plant diseases. On this background, a production level of 5,000-6,000 kg milk ha<sup>-1</sup> year<sup>-1</sup> from rather low yielding cows seems more feasible than from more high yielding cows at a lower stocking rate. It may as well be noticed that in two out of three years, grains contributed more than 40% of the energy supply which is allowed on a yearly basis according to the regulations for Norwegian organic milk production ([www.debio.no](http://www.debio.no)). In the present computations this restriction is not taken into consideration. Surpluses of grain offered for sale might, however, under the present legislations and regulations contribute to the economical sustainability of the farm.

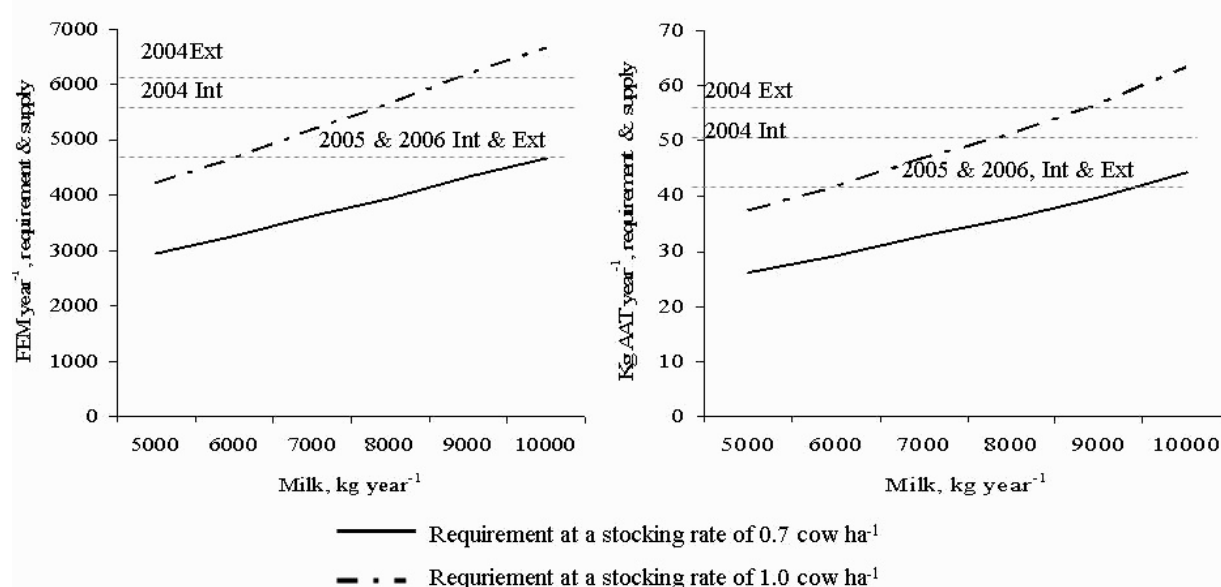


Figure 1. Total supply (shown by broken horizontal lines) of milk feed units (FEm) and amino acids absorbed in the intestine (AAT) from an organic crop rotation system under intensive (Int) and extensive (Ext) ley management in three years (2004-2006), related to animal requirements at stocking rates of 0.7 and 1.0 cow ha<sup>-1</sup> and at increasing milk yield.

## Conclusions

Under conditions as described in this study, it is possible to produce the crops needed to sustain a self sufficient dairy production of 5,000-6,000 kg milk ha<sup>-1</sup> year<sup>-1</sup>. Due to low contents of crude protein in the crops, high stocking rates with rather low yielding cows seemed to be the most feasible production strategy.

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# Effects of intensity of fertilization and cutting frequency on forage quality and diversity of permanent grassland in Central Europe in 2003-2007

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## Abstract

At six sites in the Czech Republic we evaluated long-term small-plot trials under four levels of utilization (four, three, two, and one or two cuts year<sup>-1</sup>) and four levels of fertilizer application (nil-fertilizer, PK, N<sub>90</sub>PK and N<sub>180</sub>PK). The overall average dry matter (DM) production of grasslands over four years (2003-2006) for all sites was 6.78 t ha<sup>-1</sup>, and it was significantly decreased ( $P < 0.01$ ) from 7.52 t ha<sup>-1</sup>, to 7.06 t ha<sup>-1</sup>, to 6.47 t ha<sup>-1</sup> and 6.07 t ha<sup>-1</sup> when utilized in four and three cuts in comparison with two-cut utilization. Nitrogen fertilizer application, averaged over four years, significantly increased DM production in comparison with nil-fertilized and PK-only (5.09 t and 5.47 t ha<sup>-1</sup> respectively) to 7.67 and 8.89 t DM ha<sup>-1</sup> under N<sub>90</sub>PK and N<sub>180</sub>PK treatments, respectively. Intensive (four cuts year<sup>-1</sup>) utilization of grasslands in comparison with extensive (2 cuts) utilization significantly ( $P < 0.01$ ) increased CP concentration, DM and NEL concentrations and it decreased ( $P < 0.01$ ) fibre concentration.

Keywords: permanent grassland, forage quality, cutting frequency, nitrogen fertilization, biodiversity, botanical composition

## Introduction

Permanent grasslands cover an area of 974,000 ha in the Czech Republic. However, livestock numbers have decreased from 1,236,000 cows in 1990 to 568,000 in 2006. Cutting frequency has implications for forage quality and intake. Gruber *et al.* (2000) reported that increased cutting frequency improved the voluntary intake of bulky fodder as well as daily intake of total feedstuffs (herbage + concentrates): fodder intakes were 10.4, 13.0, 15.2 kg DM at 2, 3 and 4 cuts, and there were corresponding effects on dairy production, with 11.4, 17.2 and 23.0 kg milk (FCM) per a cow per day.

## Materials and methods

Long-term small-plot trials were conducted on six permanent grassland sites in the Czech Republic, at Jevíčko, Vatín, Zubří, Liberec, Rapotín and Hladké Životice. At each site there were 16 treatments, with 4 replications, each with a 10 m<sup>2</sup> harvested-plot size. The vegetation of grasslands on the experimental stands was classified as *Arrhenatherion*. The four intensities of utilization treatments were: I<sub>1</sub> – first cut before 15 May, 4 cuts per year at 45-day intervals; I<sub>2</sub> – first cut between 16 and 31 May, 3 cuts per year at 60-day intervals; I<sub>3</sub> – first cut between 1 and 15 June, 2 cuts per year at 90-day intervals; and I<sub>4</sub> – first cut between

16 and 30 June, 1 or 2 cuts per year, second cut after 90 days. The four fertilization treatments were: F<sub>0</sub> (no fertilization), F<sub>PK</sub> (P<sub>30</sub>K<sub>60</sub>N<sub>0</sub>), F<sub>PKN90</sub> (P<sub>30</sub>K<sub>60</sub> + N<sub>90</sub>) and F<sub>PKN180</sub> (P<sub>30</sub>K<sub>60</sub> + N<sub>180</sub>). The total annual dry matter (DM) production and botanical composition of vegetation were measured at all sites. Forage quality in terms of crude protein (CP), fibre, NEL (net energy for lactation), NEV (net energy for fattening), PDIE (ingested digestive protein allowed by energy) and PDIN (ingested digestive protein allowed by nitrogen) were predicted using a NIRSystems 6500 instrument. The results presented here are averages for the harvest years 2003-2006.

## Results and discussion

The overall average grassland production (Table 1) was 6.78 t DM ha<sup>-1</sup> yr<sup>-1</sup> averaged for the 16 treatment combinations on six sites over four years (2003-2006). DM production significantly decreased ( $P < 0.01$ ) from 7.52 and 7.06 t ha<sup>-1</sup> for the 2-cuts yr<sup>-1</sup> treatments of I<sub>4</sub> and I<sub>3</sub>, to 6.07 t ha<sup>-1</sup> for the 4-cuts yr<sup>-1</sup> treatment of I<sub>1</sub>. The effect of N-fertilizer application over two years was to significantly increase ( $P < 0.01$ ) DM production compared with the nil-fertilizer and the PK-only fertilized treatments (respectively, 5.09 and 5.47 t ha<sup>-1</sup>) to 7.67 t ha<sup>-1</sup> when fertilized with 90 kg N ha<sup>-1</sup>, and 8.89 t DM ha<sup>-1</sup> when fertilized with 180 kg N ha<sup>-1</sup>. Similarly Gruber *et al.* (2000), on the basis of evaluation of long-term trials carried out at BAL Gumpenstein, reported that graded frequency of cutting decreases the yield from grasslands.

Table 1. Dry matter production and forage quality of grasslands in relation to: (A) intensity of utilization (for average levels of fertilizer application) and (B) level of fertilization (average of utilization intensities); the average of four sites and two years (2003 and 2006) of utilization.

	DM (t ha <sup>-1</sup> DM)	CP (g kg <sup>-1</sup> DM)	Fibre (g kg <sup>-1</sup> DM)	NEL (MJ kg <sup>-1</sup> DM)	NEV (MJ kg <sup>-1</sup> DM)	PDIE (g kg <sup>-1</sup> DM)	PDIN (g kg <sup>-1</sup> DM)
A (Utilization)							
I-1	6.07	173.8	213.0	5.77	5.62	85.1	103.4
I-2	6.47	154.2	235.2	5.56	5.38	82.0	90.8
I-3	7.06	127.0	271.5	5.27	5.07	77.3	74.3
I-4	7.52	122.0	287.4	5.14	4.89	74.6	71.3
Average	6.78	144.3	251.8	5.44	5.24	79.7	84.9
Dt 0.05	0.28	2.8	2.9	0.03	0.06	0.4	1.6
Dt 0.01	0.34	3.4	3.6	0.04	0.07	0.5	2.0
B (Fertilization)							
F-0	5.09	144.4	236.5	5.58	5.39	80.2	85.3
FPK	5.47	141.5	247.8	5.46	5.24	79.6	83.0
FPK N90	7.67	139.3	260.5	5.35	5.15	78.9	81.7
FPK N180	8.89	151.9	262.2	5.36	5.17	80.2	89.7
Average	6.78	144.3	251.8	5.44	5.24	79.7	84.9
Dt 0.05	0.28	2.8	2.9	0.03	0.06	0.4	1.6
Dt 0.01	0.34	3.4	3.6	0.04	0.07	0.5	2.0

From the results obtained (Table 2) it is obvious that the intensity of utilization led to a statistically highly significant decrease in the number of species in the vegetation, from 82.1 in the four-cut system of I<sub>1</sub>, to 74.0 in the extensive two-cut system of I<sub>4</sub>. This is related to a higher proportion of grasses in the vegetation; the proportion was the lowest in the I<sub>1</sub> four-cut system (54.8%) and increased to 68.1% in the I<sub>4</sub> two-cut system. The level of fertilization towards higher rates of fertilizer N was associated with a statistically significant decrease in the number of species in the vegetation, from 81.6 in the extensive nil-fertilizer treatments to 75.1 species under F<sub>PKN180</sub>. This treatment also led to statistically significant increases in the proportion of grasses in the vegetation, from 53.8 to 71.2%, and decreased the proportion of

forbs (from 36.3 to 26.6%) and legumes (from 9.8 to 2.3%). For all treatments in all years, the forage quality (Table 1) was significantly influenced by two factors. The intensive utilization ( $I_1$ ) in comparison with the extensive one ( $I_4$ ) significantly ( $P < 0.01$ ) increased CP from 122.0 to 173.8 g kg<sup>-1</sup> DM, NEL (from 5.14 to 5.77 MJ kg<sup>-1</sup> DM), NEV (from 4.89 to 5.62 MJ kg<sup>-1</sup> DM), PDIE concentration (from 74.6 to 85.1 g kg<sup>-1</sup> DM), and PDIN concentration (from 71.3 to 103.4 g kg<sup>-1</sup> DM), and it significantly ( $P < 0.01$ ) decreased the fibre concentration from 287.4 to 213.0 g kg<sup>-1</sup> DM. The level of fertilizer application, especially graded N-fertilization in comparison with the nil-fertilizer application control ( $F_0$ ) and PK fertilized ( $F_{PK}$ ) treatment, significantly ( $P < 0.01$ ) increased CP concentration from 144.4 to 151.9 g kg<sup>-1</sup> DM, NEL (from 5.58 to 5.36 MJ kg<sup>-1</sup> DM) and NEV (from 5.39 to 5.17 MJ kg<sup>-1</sup> DM). The concentration of fibre from N-fertilized treatments was increased moderately, from 236.5 to 262.2 g kg<sup>-1</sup> DM.

Table 2. The effect of utilization intensity and fertilization level on the total number of plant species at all sites, and proportions of grasses, legumes and forbs (average of 2003-2006).

Utilization intensity	Number of plant species	Grasses %	Forbs %	Legumes %	Fertilization level	Number of plant species	Grasses %	Forbs %	Legumes %
$I_1$	82.1	54.8	36.0	9.2	$F_0$	81.6	53.8	36.3	9.8
$I_2$	81.1	59.7	33.6	6.6	FPK	80.8	56.4	33.0	10.6
$I_3$	75.8	65.4	28.5	6.1	FPK N90	75.6	66.6	28.9	4.5
$I_4$	74.0	68.1	26.7	5.2	FPK N 180	75.1	71.2	26.6	2.3
Dt 0.05	3.7	1.5	1.6	0.9	Dt 0.05	3.7	1.5	1.6	0.9
Dt 0.01	4.5	1.9	1.9	1.1	Dt 0.01	4.5	1.9	1.9	1.1

## Conclusions

The overall average annual dry matter yield from grasslands over four years, for all sites, was 6.78 t ha<sup>-1</sup>, and it was significantly decreased ( $P < 0.01$ ) from 7.52 t ha<sup>-1</sup>, to 7.06 t ha<sup>-1</sup>, and to 6.47 t ha<sup>-1</sup> / 6.07 t ha<sup>-1</sup> when utilization was made using four cuts, three cuts and two cuts, respectively. Intensity of cutting frequency, and the addition of fertilizer N, resulted in changes in forage quality and botanical composition. Intensive (four cuts year<sup>-1</sup>) utilization of grasslands in comparison with extensive (two cuts) utilization significantly ( $P < 0.01$ ) increased CP concentration, DM and NEL concentrations and it decreased ( $P < 0.01$ ) fibre concentration.

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## Effects of summer sown catch crops on soil $N_{\min}$ and N removal with herbage before winter

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### Abstract

Aiming to compare the effects of summer-sown catch crops (*Lolium multiflorum* Lam., the clovers *Trifolium incarnatum* L., *Trifolium subterraneum* L., and the cruciferous crops *Brassica napus* L., *Sinapis alba* L., and *Raphanus sativus* L.) on soil mineral nitrogen ( $N_{\min}$ ) and herbage nitrogen (HN) accumulation before winter, six field experiments were carried out in the years 2004-2006 in eastern Slovenia. *L. multiflorum* and cruciferous crops effectively decreased the soil  $N_{\min}$  content. Clovers accumulated the highest amounts of HN. However, in some experiments clovers left a higher content of soil  $N_{\min}$  before winter, than did *L. multiflorum* and cruciferous crops. Fertilization with N at sowing increased the yield of herbage dry matter and HN of *L. multiflorum*, but did not increase the soil  $N_{\min}$  content before winter; this means that controlled N fertilization for forage production did not decrease the protecting role of *L. multiflorum* in soil  $N_{\min}$  depletion.

Keywords: catch crop, herbage dry matter yield, herbage nitrogen, soil mineral nitrogen

### Introduction

Crops harvested in the summer are traditionally followed by catch crops (Vos and van der Putten, 1997). Preventing N leaching, they are increasingly grown to take up soil mineral N during autumn, mild winter periods and early spring, the aim being to contribute N to the succeeding main crop in field crop rotation. In livestock production areas, the autumn yield of summer-sown catch crop can be used for forage. In such cases, N is removed from the field with forage and returned next year with organic fertilizers. In areas with unpredictable and very changeable weather, farmers must continuously adapt catch crop management to actual conditions. The management decisions about catch crops should be based on balance among unpredictable environmental conditions, environmental sustainability and farm profitability. From the environmental point of view it is important to know the potential of each individual catch crop for mineral N uptake during autumn and its potential for N accumulation in herbage. Consequently, the aim of the study was to compare the effects of summer-sown catch crops on soil mineral N content and N accumulation in herbage before winter.

### Materials and methods

Field experiments were carried out during 2004-2006 at various villages in eastern Slovenia (i.e., Experiment (Exp.) 1 in the year 2004 at the village Murski Črnci, Exp. 2 in 2005 at Lipa, Exp. 3 in 2006 at Hoče, Exp. 4 in 2006 at Rogoza, Exp. 5 in 2004 at Hoče, and Exp. 6 in 2005 at Rogoza). Treatments (catch crops) in individual Exp. (see first row in Tables 1 and 2) were arranged in a randomized block design with four replications of plots (450 m<sup>2</sup> each). Before the catch crop sowings, soil contained between 25.5 kg ha<sup>-1</sup> (Exp. 6) and 110.2 kg ha<sup>-1</sup> (Exp. 4) of mineral nitrogen ( $N_{\min}$ ) in the soil layer 0-90 cm (contents of  $N_{\min}$  in soil layers 0-30, 30-60 and 60-90 cm are presented in Tables 1 and 2). Catch crops were sown in the period 10-20 August at sowing rates: *Lolium multiflorum* 50 kg ha<sup>-1</sup>, *Raphanus raphanistrum* 20 kg ha<sup>-1</sup>, *Sinapis alba* 20 kg ha<sup>-1</sup>, *Brassica napus* 20 kg ha<sup>-1</sup>, *Trifolium incarnatum* 25 kg ha<sup>-1</sup> and *Trifolium subterraneum* 25 kg ha<sup>-1</sup>. As control, fallow land without sowing was used. Except



for the treatment *L. multiflorum* F (Table 1), which was in Exps. 1 and 2 fertilized with 60 kg N ha<sup>-1</sup> before sowing, catch crops were grown without applying any fertilizer or manure. Soil samples, which consisted of 7 subsamples from the individual plot, were taken from the plots before sowing and at the time of the herbage yield determination. N<sub>min</sub> (NH<sub>4</sub>-N and NO<sub>3</sub>-N) in soil samples was analysed according to Keeney and Nelson (1982). At the end of the growing season, the herbage yield was obtained from 7 squares (50 x 50 cm) by cutting the crop at 5 cm height and weighing. Herbage dry matter yield (HDMY) was obtained by drying samples at 70 °C for 24 h. The herbage was analysed for N content by the Kjeldahl method. Analyses of variance were carried out on herbage dry matter yield, herbage nitrogen (HN) and N<sub>min</sub> for each experiment separately. An F-ratio ≤ 0.05 was regarded as statistically significant. Comparison of treatments was made using Duncan's multiple range test.

## Results and discussion

In comparison with the control, *L. multiflorum* effectively reduced the N<sub>min</sub> content of all soil layers (Tables 1 and 2). The exception is Exp. 2 (Table 1), in which the extreme amount of precipitation leached out the majority of mineral N. The efficiency of *B. napus* in reducing the soil N<sub>min</sub> content was statistically at the same level as *L. multiflorum*. The exception was only the soil layer 30-60 in Exp. 6 (Table 2). A comparison of N accumulation in the herbage yield shows that *B. napus* accumulated more N, and that *L. multiflorum* in Exp. 4 (Table 2), and *S. alba* and *R. raphanistrum* accumulated more N in the herbage in Exp. 2, than unfertilized *L. multiflorum*. Similar soil N<sub>min</sub> contents in *L. multiflorum* and in cruciferous treatments in Exps. 2 and 4, and higher N accumulation in cruciferous crops, suggest that the cruciferous crops had been very effective in N uptake before it was leached out, or they had a non-controlled source of N, which could be only a leached N in soil layers below 90 cm. The presumption is based on findings (Thorup-Kristensen, 2001), that *L. multiflorum* is, despite its relatively shallow roots, very effective in soil N<sub>min</sub> depletion because of early N uptake, but cruciferous plants are effective because of the fast growth of very deep roots.

Table 1: The effect of *Lolium multiflorum* fertilization with N at summer sowing on soil mineral nitrogen (N<sub>min</sub>) (kg ha<sup>-1</sup>), herbage dry matter yield (HDMY) (t ha<sup>-1</sup>) and herbage nitrogen (HN) (kg ha<sup>-1</sup>) before winter.

			<i>Lolium multiflorum</i> 0 <sup>2</sup>	<i>Lolium multiflorum</i> F	<i>Sinapis alba</i>	<i>Raphanus raphanistrum</i>	Control
Exp. 1	N <sub>min</sub> 0-30	(36.4) <sup>1</sup>	17.5 <sup>b*</sup>	19.7 <sup>b</sup>			32.2 <sup>a</sup>
	N <sub>min</sub> 30-60	(12.6)	10.4 <sup>b</sup>	11.3 <sup>b</sup>			29.7 <sup>a</sup>
	N <sub>min</sub> 60-90	(8.2)	9.4 <sup>b</sup>	10.9 <sup>b</sup>			17.8 <sup>a</sup>
	HDMY		1.67 <sup>b</sup>	2.74 <sup>a</sup>			-
	HN		40.5 <sup>b</sup>	70.9 <sup>a</sup>			-
Exp. 2	N <sub>min</sub> 0-30	(23.1)	5.3	5.2	6.2	5.3	7.7
	N <sub>min</sub> 30-60	(10.7)	5.8	6.8	6.2	6.0	6.8
	N <sub>min</sub> 60-90	(8.3)	5.5 <sup>b</sup>	6.5 <sup>ab</sup>	5.9 <sup>b</sup>	5.8 <sup>b</sup>	7.5 <sup>a</sup>
	HDMY		1.31 <sup>b</sup>	2.20 <sup>a</sup>	2.41 <sup>a</sup>	1.66 <sup>b</sup>	0.68 <sup>c</sup>
	HN		27.1 <sup>b</sup>	50.6 <sup>a</sup>	50.3 <sup>a</sup>	36.6 <sup>b</sup>	12.9 <sup>c</sup>

<sup>1</sup> Soil mineral N content at sowing.

<sup>2</sup> *Lolium multiflorum* 0, non-fertilized at sowing; *Lolium multiflorum* F, fertilized with 60 kg N ha<sup>-1</sup> at sowing; other treatments were also non-fertilized with N at sowing.

\*Numbers followed by different letters within a row are significantly different ( $P \leq 0.05$ ).

*T. subterraneum* is not a common catch crop in Central Europe; however it was included in experiments because of its promising results in our previous work (Kramberger *et al.*, 2006). In Exps. 3 and 5 (Table 2), *T. subterraneum* depleted soil N<sub>min</sub> to the statistically same level as *L. multiflorum*, but in the soil layer 0-30 cm of Exp. 6 and in all three soil layers of Exp. 4,

the values of  $N_{\min}$  content were higher for *T. subterraneum* than *L. multiflorum*. The values of soil  $N_{\min}$  content were higher also in treatment of *T. incarnatum* in Exps. 3 and 4. The clovers accumulated significantly the highest amounts of HN (Table 2), which can be attributed also to symbiotic N fixation. A part of the accumulated N was obviously released into the soil in Exps. 3, 4 and 6.

Table 2: The effect of different catch crops, sown in summer, on soil mineral nitrogen ( $N_{\min}$ ) ( $\text{kg ha}^{-1}$ ), herbage dry matter yield (HDMY) ( $\text{t ha}^{-1}$ ) and herbage nitrogen (HN) ( $\text{kg ha}^{-1}$ ) before winter.

		<i>Lolium multiflorum</i>	<i>Trifolium subterraneum</i>	<i>Trifolium incarnatum</i>	<i>Brassica napus</i>	Control
Exp. 3	$N_{\min}$ 0-30 (37.1)*	8.7 <sup>b*</sup>	4.0 <sup>b</sup>	19.1 <sup>a</sup>	5.3 <sup>b</sup>	19.5 <sup>a</sup>
	$N_{\min}$ 30-60 (26.2)	9.4 <sup>c</sup>	13.0 <sup>bc</sup>	37.9 <sup>a</sup>	8.4 <sup>c</sup>	18.6 <sup>b</sup>
	$N_{\min}$ 60-90 (15.1)	11.0 <sup>b</sup>	13.0 <sup>b</sup>	20.0 <sup>a</sup>	9.7 <sup>b</sup>	19.8 <sup>a</sup>
	HDMY	1.4 <sup>d</sup>	2.3 <sup>b</sup>	2.9 <sup>a</sup>	1.7 <sup>c</sup>	
	HN	17.4 <sup>d</sup>	81.1 <sup>b</sup>	116.9 <sup>a</sup>	33.2 <sup>c</sup>	
Exp. 4	$N_{\min}$ 0-30 (54.1) <sup>1</sup>	12.6 <sup>c</sup>	23.3 <sup>b</sup>	25.1 <sup>b</sup>	14.8 <sup>c</sup>	33.7 <sup>a</sup>
	$N_{\min}$ 30-60 (42.3)	14.8 <sup>c</sup>	25.1 <sup>b</sup>	33.7 <sup>a</sup>	23.3 <sup>b</sup>	12.6 <sup>c</sup>
	$N_{\min}$ 60-90 (13.8)	10.3 <sup>b</sup>	26.5 <sup>a</sup>	14.9 <sup>b</sup>	13.5 <sup>b</sup>	30.4 <sup>a</sup>
	HDMY	1.1 <sup>b</sup>	1.3 <sup>b</sup>	1.9 <sup>b</sup>	3.8 <sup>a</sup>	-
	HN	14.9 <sup>c</sup>	58.8 <sup>b</sup>	89.4 <sup>a</sup>	67.1 <sup>b</sup>	-
Exp. 5	$N_{\min}$ 0-30 (22.7)	18.3 <sup>b</sup>	21.3 <sup>ab</sup>		15.7 <sup>b</sup>	29.7 <sup>a</sup>
	$N_{\min}$ 30-60 (36.8)	11.0 <sup>b</sup>	13.2 <sup>b</sup>		9.8 <sup>b</sup>	18.3 <sup>a</sup>
	$N_{\min}$ 60-90 (9.6)	12.9 <sup>b</sup>	12.0 <sup>b</sup>		15.5 <sup>ab</sup>	20.7 <sup>a</sup>
	HDMY	2.1 <sup>b</sup>	2.8 <sup>a</sup>		1.9 <sup>b</sup>	-
	HN	54.7 <sup>b</sup>	102.4 <sup>a</sup>		51.0 <sup>b</sup>	-
Exp. 6	$N_{\min}$ 0-30 (10.8)	16.4 <sup>c</sup>	22.5 <sup>bc</sup>		23.2 <sup>b</sup>	29.6 <sup>a</sup>
	$N_{\min}$ 30-60 (7.7)	12.5 <sup>ab</sup>	10.2 <sup>ab</sup>		8.6 <sup>b</sup>	13.9 <sup>a</sup>
	$N_{\min}$ 60-90 (7.0)	7.7	7.5		7.3	7.9
	HDMY	0.6 <sup>b</sup>	1.11 <sup>a</sup>		0.4 <sup>b</sup>	0.4 <sup>b</sup>
	HN	15.5 <sup>b</sup>	40.1 <sup>a</sup>		10.3 <sup>b</sup>	8.3 <sup>b</sup>

<sup>1</sup>Soil mineral N content at sowing.

\*Numbers followed by different letters within a row are significantly different ( $P \leq 0.05$ ).

As we were aiming at soil  $N_{\min}$  depletion, catch crops were not fertilized with N (except fertilized *L. multiflorum* in Exps. 1 and 2). Despite this, the amounts of HDMY before winter were, in treatments with clovers, and sometimes also in treatments with cruciferous crops or *L. multiflorum*, large enough for use as forage. *L. multiflorum* fertilization with N at sowing (Table 1) did not increase soil  $N_{\min}$  content before winter. However, it significantly increased HDMY and HN, suggesting that controlled N fertilization for autumn forage does not decrease the protecting role of *L. multiflorum* in soil  $N_{\min}$  depletion.

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# Long-term changes of $^{15}\text{N}$ natural abundance of plants and soil in grassland

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## Abstract

The natural abundance of  $^{15}\text{N}$  in soils and plants was analysed in a long-term lysimeter determining the suitability of  $\delta^{15}\text{N}$  as a marker of nitrogenous loss rates. Organic (cattle slurry) and mineral (calcium ammonium nitrate) fertilizers were applied in rates between 0 and 480 kg N ha<sup>-1</sup>yr<sup>-1</sup>. The mean  $\delta^{15}\text{N}$  value of the mineral fertilizer was  $-1 \pm 0.2\text{‰}$  and that of the organic fertilizer was  $8.9 \pm 0.5\text{‰}$ .  $\delta^{15}\text{N}$  values of soil-N and plant material increased with the amounts of applied N-fertilizers. Further, samples receiving organic fertilizer were more enriched in  $\delta^{15}\text{N}$  than samples with mineral N, reflecting higher  $\delta^{15}\text{N}$  values of organic compared to mineral fertilizer. The  $\delta^{15}\text{N}$  values of top soil samples increased from  $1.7 \pm 0.2\text{‰}$  to  $6 \pm 0.2\text{‰}$  and  $\delta^{15}\text{N}$  values of plant material increased from  $1.2 \pm 0.6\text{‰}$  to  $4.4 \pm 0.3\text{‰}$ , when N applications were high. Obviously, higher  $\delta^{15}\text{N}$  values were attributable to inefficient N usage, i.e. nitrogenous losses, due to  $^{15}\text{N}$  fractionation during  $\text{NH}_3$  volatilization.

Keywords: N-fertilizer application,  $\delta^{15}\text{N}$ , natural abundance, N-efficiency

## Introduction

High amendments of nitrogenous fertilizers stimulate biomass production and increase the N content in plant material. However, any N fertilizer application is accompanied by undesirable side effects related to N losses through ammonia volatilization,  $\text{NO}_3^-$  leaching or nitrogenous trace gas emissions (Dittert *et al.*, 1998). N transformation within the ecosystem leads to N isotope fractionation. Such  $^{15}\text{N}$  fractionations may reach up to 35‰ (Högberg, 1997). According to the Rayleigh model (Robinson, 2001) all products that are emitted to the environment ( $\text{NH}_3$ ,  $\text{N}_2\text{O}$ ,  $\text{NO}$ ,  $\text{N}_2$ ,  $\text{NO}_3$ ) are generally  $^{15}\text{N}$  depleted and hence, the remaining natural N-sources are enriched in  $^{15}\text{N}$  (Högberg, 1997). Such enrichment might be a promising signature to reconstruct N losses due to management in different compartments of an agricultural farm (Watzka *et al.*, 2006). We hypothesize that the long-term use of different forms of N fertilizer application can be traced back by characteristic  $\delta^{15}\text{N}$  values in soils and plants. We further hypothesized that elevated  $\delta^{15}\text{N}$  values within appropriate N-pools indicate N-surplus leading to N losses to the environment. We also explored if the  $\delta^{15}\text{N}$  values of plant biomass is indirectly influenced by floristic composition as specific isotope discrimination might differ among plant species.

## Materials and methods

Plant samples were collected from a 22-year-old field lysimeter experiment at the Research Station Rengen (Eifel Mountains, Germany). The experiment was performed in three different treatments in a randomized complete block design, with each 3 blocks consisting of 5 permanent grassland plots (2 x 6 m). N was either applied in organic fertilizer (cattle slurry), mineral fertilizer (calcium ammonium nitrate) or as a mixture of both. Four cuts per season were taken. Pooled samples of plant biomass were collected before the second and fourth cut. A total of 10-50 plant leaves (depending on plant species, weight and availability) were therefore cut 1 cm above ground. To account for species-specific isotope composition, *Lolium perenne* L., *Festuca rubra* L., *Trifolium repens* L., *T. pratense* L. and *Taraxacum officinale*

Web. were sampled. Bulk soil samples ( $n = 10$ ) were taken from each plot concurrently and were attributed to subsamples from 0-5 cm, 5-10 cm and 10-30 cm soil depth. Total N content as well as natural  $^{15}\text{N}$  abundances in all samples were determined by dry combustion using an elemental analyser (NA 1108, Carlo Erba, Milan, Italy) coupled with a continuous-flow isotope ratio mass spectrometer (Delta Plus, ThermoFinnigan, Bremen, Germany). All isotope values were expressed relative to the international standards for isotope composition (IAEA, Vienna, Austria). Statistical analysis was performed separately for each experiment using SPSS 14.0 software. Differences between fertilizer treatments were analysed by one-way analysis of variance (ANOVA, 95% confidence interval) followed by a multiple range test (Scheffé *post hoc* test). If homogeneity of variance was missing, Dunnett's T3 was calculated.

## Results and discussion

Bulk soil N contents hardly changed with increased N fertilizer supply (data not shown), suggesting that N losses in soils increased as a result of increased fertilizer application. Indeed, the  $\delta^{15}\text{N}$  values of both soils and harvested plant biomass increased with the amount of applied N fertilizer. Soil  $\delta^{15}\text{N}$  values ranged from  $1.8 \pm 0.2\text{‰}$  to  $6.0 \pm 0.2\text{‰}$  in the upper horizons and  $3.5 \pm 0.5\text{‰}$  to  $5.5 \pm 0.6\text{‰}$  in deeper soil layers (Figure 1). The results support the hypothesis, that increasing N applications were reflected by increased soil  $\delta^{15}\text{N}$  values.

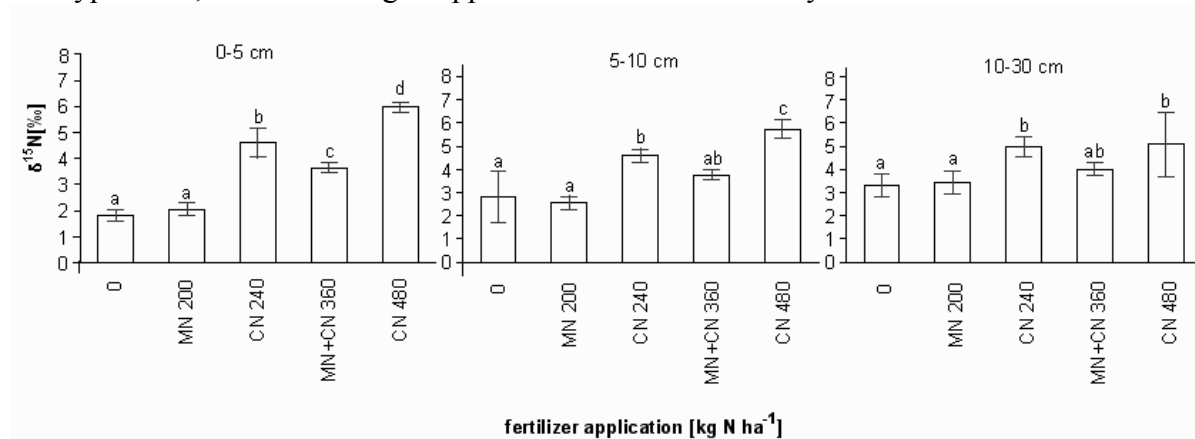


Figure 1. Mean  $\delta^{15}\text{N}$  abundances of soils in relation to amount of fertilizer N applied as cattle slurry (CN) mineral (MN) and mixed fertilizers (MN + CN) at three soil depths in 2006 ( $n = 6$ ). Values with the same letters are not significantly different (one-way ANOVA,  $P < 0.05$ ). Error bars represent the 95% confidence interval of the means.

The  $\delta^{15}\text{N}$  value for the added mineral fertilizer averaged  $-1 \pm 0.2\text{‰}$ , hence the increased  $\delta^{15}\text{N}$  value with increased inorganic fertilizer application was not due to the fertilizer signal itself. It is likely that this shift was attributable to transformation of fertilizer-N and its associated isotope fractionation (Handley and Raven, 1992; Yoneyama, 1996; Högberg, 1997; Dawson, 2002). All soil transformations leading to N losses result in considerable fractionations, especially during  $\text{NH}_3$  volatilization (29‰), denitrification (0-33‰) and nitrification (15-35‰). Accordingly, products of N fractions lost to the environment ( $\text{NH}_3$ ,  $\text{N}_2\text{O}$ ,  $\text{NO}$ ,  $\text{N}_2$ ,  $\text{NO}_3$ ) are  $^{15}\text{N}$  depleted while the residual N-sources were enriched in  $^{15}\text{N}$  (Handley and Raven, 1992; Yoneyama, 1996).

Similarly to soils, plant  $\delta^{15}\text{N}$  increased with increasing amount of applied N depending on the N form. The above ground total biomass  $\delta^{15}\text{N}$  values ranged from  $-1.2 \pm 0.6\text{‰}$  to  $4.4 \pm 0.3\text{‰}$  (Figure 2). This finding suggests that major N signals from the fertilizer N were recovered in the plants. Further, plant  $\delta^{15}\text{N}$  values were correlated with the corresponding top soils for all

species except for *Trifolium repens* L. and *Trifolium pratense* L.. Distinctions in  $\delta^{15}\text{N}$  among plant species may reflect differences in plant growth, resource acquisition strategies or varieties in  $\delta^{15}\text{N}$  of soil N pools for which they compete (Nadelhoffer *et al.*, 1996).

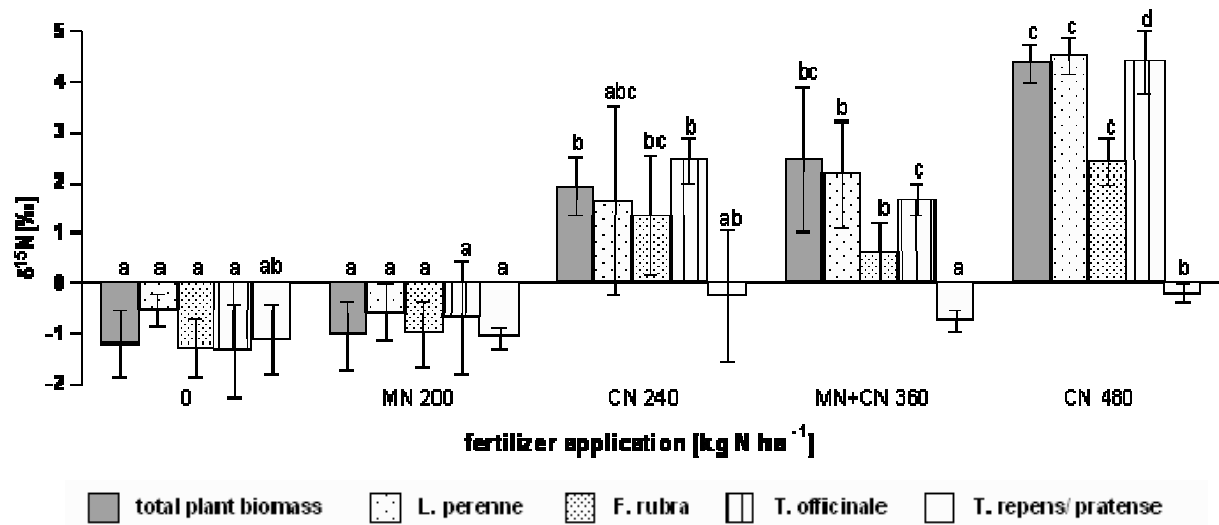


Figure 2. Mean  $\delta^{15}\text{N}$  abundances of above ground plant biomass in relation to amount of applied fertilizer N (for abbreviations see Figure 1). Five plant species and additionally one total plant biomass were collected in 2006. Values with the same letters are not significantly different (one-way ANOVA,  $P < 0.05$ ). Error bars represent the 95% confidence interval of the means.

## Conclusions

The  $\delta^{15}\text{N}$  values of soils and above-ground biomass were highly correlated with the fertilizer management. Thus, not only quantity but also chemical form of applied N-fertilizer was reflected by the variations in  $\delta^{15}\text{N}$  abundance of each component. Despite the large variability of  $\delta^{15}\text{N}$  values found in different compartments of the ecosystems, results thus clearly showed that it is possible to use the  $\delta^{15}\text{N}$  natural abundance technique to track hot spots of inefficient N usage in a 22-year-old experiment.

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# Alternative feeding regimes to ewes in organic farming in northern Norway

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## Abstract

With a demand of 100% organic feed in organic sheep production the provision of feed is a challenge for many organic farmers in Norway. Organic concentrates are expensive compared with conventional concentrates and in northern Norway the climate limits the production of grain and protein-rich feeds. An experiment in organic lamb meat production in northern Norway was launched in 2002. Norwegian sheep received different indoor feeding regimes during late gestation and early lactation. The aim of the experiment was to compare a traditional diet based on grass silage and commercial concentrate, with a diet based on grass silage, potatoes and fishmeal, without changing the animal performance. Ewe-weight changes between the control diet and the experimental diet were not different (2.0 and 1.5 kg, respectively). Neither was the lambs' average daily gain different between treatments (291 and 290 g d<sup>-1</sup>, respectively). It is concluded that a supplement diet containing organic potatoes and fishmeal can be used as an alternative to a diet containing traditional organic commercial concentrate to ewes during late gestation and early lactation.

Keywords: concentrate, potato, fish meal, lamb weight

## Introduction

In Norway, the farming systems have great challenges due to different climate zones. In northern Norway the grazing season is short and housing is needed for the grazing animals. The sheep are kept indoors for about 200 days from the start of mating of the ewes in the autumn until after lambing in the spring. The ewes are mainly fed grass silage and small amounts of supplements during the low pregnancy period. After lambing the sheep are released for spring and summer grazing. The Norwegian set of rules for organic farming is close to the European rules: a demand for 100% organic feed to ruminants of which 50% must have been produced on the farm. At least 60% of the diet (DM) must be roughage and a maximum of 40% can be supplements based on grain on an annual basis. Traditionally, in northern Norway the agricultural system is based on grass production. Potatoes are a common crop grown with a high yield of good quality. Potatoes not suitable for human consumption, due to size and physical damage, have potential as animal feed. When feeding potatoes it is important that the potatoes are clean and free from sprouts. Green potatoes and sprouts contain solanine which in some cases can be lethal (Morris and Lee, 1984). Commercial organic concentrates are already on the market. However, the concentrates are expensive. Protein-rich feeds like rape, peas and lupins are difficult to grow in northern Norway due to the climate. In Norway, fish meal is still allowed to be used in an organic production and locally produced fish meal is available. Fish meal is rich in protein and is an alternative protein source. The purpose of this experiment was to investigate potatoes and fish meal as a supplement to ewes during late gestation and early lactation in northern Norway.

## Materials and methods

A feeding experiment with sheep (Old Norwegian short tailed landrace) was initiated at Tjøtta Rural Development Centre (65°50'N, 12°28'E) in northern Norway in 2002. The experimental period lasted from eight weeks prior to lambing until slaughter in the autumn. Eight weeks prior to average lambing (March 20) 42 ewes were randomly selected, weighed and divided into 6 pens with 7 ewes in each pen. Three pens received the control feed (grass silage and concentrate) and three pens received the experimental feed (grass silage, potato and fish meal). Ewes with newborn lambs (average date of birth, May 4) were put in individual pens for about 48 hours and then merged with the ewes and lambs from the same feeding regime. When the lambs were nine to twelve days old, ewes and lambs were weighed (indoor weighing) and released onto cultivated pastures for spring grazing. Ewes and lambs were collected for spring weighing (June 14) and released onto a mountain pasture until September 7th. Then the lambs were collected and weighed (autumn weight) before transported back to Tjøtta.

The ewes were offered grass silage *ad libitum* and supplements twice every day during the indoor feeding period. The amount of supplements offered to the ewes was determined by using the Norwegian recommendations (Nedkvitne, 1987). Supplement content in the rations was balanced with respect to energy. This caused small but not significant differences in the amount of crude protein in supplements between diets.

Data were analysed as a completely randomized GLM (General Linear Model) design using Minitab Statistical Software. When analysing lambs average daily weight gain, initial live weight was used as a covariate.

## Results and discussion

No differences between feeding regimes were found on feed intake (Table 1) before or after lambing. The ewes gained weight during the indoor feeding period (1.5 kg in control pens and 0.5 kg in experimental pens; not significant) and lost weight during spring grazing (1.2 and 2.1 kg respectively; not significant). Potatoes contain a high level of water (70-75%) and are rich in starch but very low in fibre, protein and vitamins A and D. Potatoes used as a feed for ruminants are characterized as a supplement. Since the level of crude protein is low it is important to add a feed with high levels of protein together with potatoes. Fish meal has been shown to be less degradable in the rumen than other commonly used protein supplements (Gonzales *et al.*, 1979). Addition of fish meal to the diet of ewes in early lactation is more likely to increase amino acid supply to the intestines and so to protein output in milk, than additions of more degradable supplements. Our findings are in agreement with this, since the change of body weight in the ewes not was affected by the protein source.

We did not find any differences on lamb performance between the feeding regimes, either on average daily weight gain during the indoor feeding period or during the grazing season (Table 1). Lambs from the experimental diet tended ( $P = 0.1$ ) to have a higher live weight at autumn. However, this was not reflected in slaughter weight ( $P = 0.5$ ), EUROP-classification ( $P = 0.7$ ) or EUROP-fat-classification ( $P = 0.3$ ). It was not expected to find any differences on lamb performance since the ewe performance was not different between the feeding regimes.

The results (Table 1) show that feeding potatoes and fish meal as a supplement to ewes during late gestation and early lactation was a good substitute for commercial organic concentrate.



Table 1. Feed intake per ewe per day in kg dry matter (DM), energy (SFU d<sup>-1</sup>), metabolic energy (MJ d<sup>-1</sup>) and crude protein (g d<sup>-1</sup>) before and after lambing. Ewes average start weight (March 20), indoor weight (released onto spring pasture), spring weight (released on to mountain pasture). Lambs average weight at birth, indoor, spring and autumn (collected from the mountain pasture). Lambs average daily gain (ADG) indoors, on spring and summer pasture (g d<sup>-1</sup>).

	Control	Experimental	SEM	P
No of pens	3	3		
<i>Feed intake before lambing, animal d<sup>-1</sup></i>				
Grass silage, kg DM	1.8	1.8	0.1	0.9
Concentrate, kg DM	0.49			
Potato, kg DM		0.42		
Fish meal, kg DM		0.09		
Total E, SFU	1.9	2.0	0.1	0.9
Total ME, MJ	23.1	23.4	0.1	0.8
Total CP, g	312	317	5	0.9
<i>Feed intake after lambing, animal d<sup>-1</sup></i>				
Grass silage, kg DM	1.7	1.7	0.1	0.9
Concentrate, kg DM	0.78			
Potato, kg DM		0.56		
Fish meal, kg DM		0.17		
Total E, SFU	2.1	2.1	0.1	0.9
Total ME, MJ	25.7	25.3	0.2	0.8
Total CP, g	337	355	15	0.7
<i>Ewe performance</i>				
Weight start, kg	60.5	64.5		
Weight indoor, kg	62.5	65.0	2.1	0.7
Weight spring, kg	61.3	62.9	2.4	0.6
<i>Lamb performance</i>				
No of lambs	24	25		
Birth weight, kg	4.8	5.3	0.2	0.3
Weight indoor, kg	8.6	9.1	0.3	0.4
Weight spring, kg	13.9	15.4	0.6	0.1
Weight autumn, kg	40.4	43.3	1.5	0.1
ADG indoor, g d <sup>-1</sup>	291	290	21	0.4
ADG spring pasture, g d <sup>-1</sup>	242	256	13	0.5
ADG summer pasture, g d <sup>-1</sup>	283	309	10	0.3

## Conclusions

It is concluded that organic potatoes and fish meal can be used as an alternative to commercial concentrates to ewes during late gestation and early lactation without changing the ewe or lamb performance.

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## Effect of different management systems on sown meadows

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### Abstract

The aim of this study was to compare Italian ryegrass-red clover (*Lolium multiflorum* Lam.-*Trifolium pratense* L.) sown meadows established under conventional (CMS) and organic management systems (OMS). Experiments were conducted simultaneously at two different locations using field-scale (1.5 ha) or experimental (90 m<sup>2</sup>) plots, with a randomized block design having four replications of each treatment. Meadows were sown in autumn 2006 at seed rates of 22:12 kg ha<sup>-1</sup> of Italian ryegrass: red clover. The NPK fertilizer dose applied for CMS was 190, 120, 120 kg ha<sup>-1</sup> yr<sup>-1</sup>, for N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O, respectively. Sown meadows growing under OMS were fertilized with cattle manure containing 2.7 kg N m<sup>-3</sup> and high concentrations of P, K and Mg, using a dose of 45 m<sup>3</sup> ha<sup>-1</sup> yr<sup>-1</sup> distributed in three applications. Differences between CMS and OMS were measured in terms of production, botanical composition, nutritive values and ensilability characteristics in each cut. Preliminary results showed considerable changes in botanical composition due to management system ( $P < 0.001$ ). The annual herbage dry matter yield was higher ( $P < 0.001$ ) for CMS, but with no significant differences in crude protein or metabolizable energy ( $P > 0.05$ ). Ensilability characteristics were better in samples from the OMS treatments due to higher dry matter and water soluble carbohydrate contents, compared with CMS samples.

Keywords: organic management system, conventional management system, productivity, botanical composition, nutritive value, ensilability characteristics

### Introduction

In order to reduce animal production costs and to improve environmental conditions, there is a growing interest in lower-input pasture systems. In this context, having an appropriate proportion of clovers (*Trifolium* spp.) in meadows and pastures guarantees high nutritive value of forage for ruminants as a result of increased contents of protein and minerals, as well as improvement of forage digestibility and palatability. Clovers also contribute to the nitrogen (N) input via symbiotic fixation, and this supply of N plays an important role both in sustainable and low-input systems of feed production, and it is particularly important for organic farms as they usually have a negative N balance. The application of mineral fertilizers in conventional management systems (CMS) changes the botanical composition, and hence the nutritive value of the herbage. Nitrate fertilization affects negatively two of the most important legumes present in meadows in northern Spain (red clover and white clover). However, it is assumed that P and K fertilizers improve legume development (García *et al.*, 2005). Unlike CMS, organic management systems (OMS) have to rely on efficient nutrient cycling within the farm to maintain soil fertility and sustainable production. The purpose of this work was to compare, at experimental and field level, the effect of organic and conventional management systems on productivity, botanical composition and herbage quality of Italian ryegrass-red clover sown meadows.

## Materials and methods

Experiments were conducted in the North-west of Spain at two different locations simultaneously (a coastal site and an inland valley) on field plots of 1.5 ha or experimental plots of 90 m<sup>2</sup>, using a randomized block design with four replications of each treatment. All plots were sown in October 2006 using a seed rate of 22:12 kg ha<sup>-1</sup> for Italian ryegrass (cv. Ansyl) and red clover (cv. Violetta), respectively. The NPK fertilizer dose applied for the CMS was 190, 120, 120 kg ha<sup>-1</sup> yr<sup>-1</sup>, for N (ammonium nitrate), P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O, respectively. Meadows managed organically (to European regulation 2092/1991) were fertilized using cattle manure that contained 2.7, 1.4, 5.2, and 0.7 kg m<sup>-3</sup> of N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O and MgO, respectively, at 45 m<sup>3</sup> ha<sup>-1</sup> yr<sup>-1</sup> distributed in three applications (one at sowing and two in spring). Meadows were cut six times during the first year, depending on climatic conditions. For each cut the total yield (Mg DM ha<sup>-1</sup>) was determined. Forage samples were also analysed for chemical composition. Dry matter (DM) of samples was determined by drying in an air-forced oven at 102 °C for 24 h. Nutritive values were analysed by near infrared reflectance spectroscopy (NIRS) and the metabolizable energy (ME) was estimated (ARC, 1980). Samples from spring cuts (for silage) were also analysed for ensilability characteristics (De la Roza-Delgado *et al.*, 2004). In addition, for each cut, botanical composition of forages was determined by hand separating into different fractions (sown species, other species and dead matter), and expressed as percentage of total DM. All data were analysed using SAS (1999).

## Results and discussion

Evaluation of botanical composition is shown in Table 1. Under both management systems, Italian ryegrass decreased significantly ( $P < 0.001$ ) during summer, probably due to the high rainfall in this period. In the OMS red clover had a competitive advantage ( $P < 0.001$ ). Mineral fertilization limited significantly the legumes grown in CMS.

Table 1. Changes in botanical composition of Italian ryegrass-red clover sown meadows under different management systems (mean values are percentages of total DM).

	Conventional management system						Organic management system						s.e.	Significance		
	1C	2C	3SC	4SC	5C	6C	1C	2C	3SC	4SC	5C	6C		C	M	C*M
Italian ryegrass	69	63	95	90	60	3	68	68	87	52	30	1	1.65	***	***	**
Red clover	8	1	2	5	33	78	6	2	9	41	68	88	1.32	***	***	***
Other species	21	30	1	2	5	3	22	24	2	3	1	1	0.97	***	ns	ns
Dead material	2	6	2	3	2	16	4	6	2	4	1	10	0.29	***	ns	**

\*, \*\* and \*\*\* Significant at 0.05, 0.01 and 0.001% levels respectively. ns  $P > 0.05$ ; s.e.: mean standard error. C: Cut; SC: Silage cut; M: Management.

The annual yield was higher ( $P < 0.001$ ) for CMS, with significant differences between size of plots ( $P < 0.001$ ). These marked differences between the field plots and experimental plots (11.2 vs. 19.1 Mg DM ha<sup>-1</sup> y<sup>-1</sup>) may have been caused by different harvesting systems. The significantly greater amount of clover in the OMS, compared with CMS, resulted in a lower fibre content ( $P < 0.001$ ) due to higher proportion of leaves and stems (Table 2), but without significant differences in ash, crude protein (CP) and metabolizable energy (EM) ( $P > 0.05$ ). Results in Table 3 show the ensilability characteristics under both management systems. The DM and water soluble carbohydrate contents were higher in OMS than CMS (174 vs. 161 g kg<sup>-1</sup>;  $P > 0.05$ ), and (150 vs. 119 g kg<sup>-1</sup>DM;  $P < 0.05$ ). There were no significant differences in buffering capacity between OMS and CMS (402 vs. 397 meq NaOH kg<sup>-1</sup>DM;  $P > 0.05$ ). As a result, the fermentability coefficient (FC; Weissbach, 1999) appeared higher for OMS but without significant differences. Significant differences for FC were observed between different sized plots (27 vs. 16;  $P < 0.01$ ) of the field and experimental plots.

Table 2. Annual yield, nutritive value and energy content of Italian ryegrass-red clover sown meadows under different management systems.

	Conventional management system	Organic management system	s.e.	Significance					
				C	M	S	C*M	C*S	M*S
Annual yield (Mg DM ha <sup>-1</sup> y <sup>-1</sup> )	17.7	13.1	0.04	***	***	***	***	***	***
Ash (g kg <sup>-1</sup> DM)	103	103	0.13	***	ns	***	ns	***	ns
CP (g kg <sup>-1</sup> DM)	176	172	0.15	***	ns	***	***	***	ns
NDF (g kg <sup>-1</sup> DM)	471	437	0.39	***	***	***	ns	***	ns
ADF (g kg <sup>-1</sup> DM)	268	251	0.16	***	***	*	*	***	ns
ME (MJ kg <sup>-1</sup> DM)	10.5	10.4	0.03	***	ns	***	***	***	*

\*, \*\* and \*\*\* Significant at 0.05, 0.01 and 0.001% levels respectively. ns  $P > 0.05$ ; s.e.: mean standard error; C: Cut; M: Management; S: Size plot; DM: Dry matter; CP: Crude protein; NDF: Neutral detergent fibre; ADF: Acid detergent fibre; ME: Estimated metabolizable energy.

Table 3. Ensilability characteristics of Italian ryegrass-red clover sown meadows under different management systems.

	Conventional management system		Organic management system		s.e.	Significance				
	3SC	4SC	3SC	4SC		C	M	S	C*M	C*S
DM (g kg <sup>-1</sup> )	137	186	161	188	0.83	*	ns	**	ns	ns
WSC (g kg <sup>-1</sup> DM)	130	108	168	132	0.59	*	*	***	ns	ns
BC (meq NaOH kg <sup>-1</sup> DM)	360	434	402	401	1.95	ns	ns	*	ns	ns
FC	18	23	21	24	1.16	ns	ns	*	ns	ns

\*, \*\* and \*\*\* Significant at 0.05, 0.01 and 0.001% levels respectively. ns  $P > 0.05$ ; s.e.: Mean standard error; C: Cut; SC: Silage cut; M: Management; S: Size plot; DM: Dry matter; WSC: Water soluble carbohydrates; BC: Buffering capacity; FC: Fermentability coefficient.

## Conclusions

Preliminary results show that the annual DM yield is higher under the CMS with high inputs of mineral fertilization ( $P < 0.001$ ). However, this mineral fertilization limits significantly the legumes grown and induces considerable changes in botanical composition. Ensilability characteristics were better in samples growing under OMS, with higher water soluble carbohydrate contents than samples from the CMS treatments.

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# The effect of cattle-slurry electroflotation on gaseous emissions from grasslands

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## Abstract

The climatic characteristics of the Basque Country (northern Spain) provide favourable conditions for the growth of grasslands, with dairy cattle the major farming activity in the area. ADE BIOTEC S.L. is developing a process called electroflotation, with the aim of reducing the volume of slurries from intensive livestock farms. This process gives as a final result two different subproducts: a solid phase and a liquid phase. These subproducts can be applied as organic fertilizers. Both the solid and the liquid phases reduced NH<sub>3</sub> volatilization rates and NO losses, without any effect on N<sub>2</sub>O and CO<sub>2</sub> emissions.

Keywords: ammonia volatilization, carbon dioxide, electroflotation, nitric oxide, nitrous oxide

## Introduction

The intensification of farms is leading to serious environmental risks due to the large production of manures and slurries and their subsequent inefficient management. Their application as fertilizer involves N losses that can lead to pollution and difficulties for intensive farmers to comply with legislation on greenhouse gas emissions (Kyoto Protocol) ammonia (Gothenburg Protocol) and water quality (EU Nitrate Directive). An 'electroflotation' process developed by ADE BIOTEC S.L. gives two different subproducts. The liquid phase can be used as 'fert-irrigation' water on grassland, while the solid phase can be applied directly as an organic amendment. The objective of this work was to assess the polluting effect of the two phases obtained by electroflotation, by determining their effect on gaseous emissions after their application on grasslands in comparison with untreated slurry.

## Materials and methods

In spring, an experiment was established in a cut grassland (on a poorly drained clay loam) in Derio (Basque Country). The region has a temperate climate with a typical annual mean temperature of 12 °C and a high rainfall, 1,200 mm yr<sup>-1</sup>. A typical permanent pasture seed mixture (*Lolium perenne* L. var. Herbus, 60%; *Lolium hybridum* L. var. Texi, 32%; *Trifolium repens* L. var. Huia, 8%) was sown at a density of 40 kg seeds ha<sup>-1</sup> in the previous autumn. The original untreated cow slurry was obtained from a concrete storage pit on a dairy farm. Untreated slurry as well as solid phase and liquid phase were applied on the surface at a rate of 70 kg NH<sub>4</sub><sup>+</sup>-N ha<sup>-1</sup>. A treatment with no fertilizer was also included as a control. Four microplots (0.03 m<sup>2</sup>) per treatment were established. The characteristics of the original untreated slurry, the solid phase and the liquid phase, were analysed just before their application as shown in Table 1.

Nitrous oxide and carbon dioxide emissions were measured using a closed air circulation technique in conjunction with a photoacoustic infrared gas analyser during a 40-minute period (Menéndez *et al.*, 2006).

Table 1. Characteristics of the untreated slurry, the solid phase and the liquid phase applied in both experiments.

	Experiment 1		
	Untreated Slurry	Solid phase	Liquid phase
Total N, g kg <sup>-1</sup> FW	0.50	0.18	0.07
NH <sub>4</sub> <sup>+</sup> -N, g kg <sup>-1</sup> FW	0.24	0.06	0.06
C, g kg <sup>-1</sup>	3.38	1.71	0.51
C:N	6.76	9.50	7.28
DM, g kg <sup>-1</sup>	11.50	4.83	1.47
pH	8.34	6.98	7.46

Nitric oxide emissions were measured just before N<sub>2</sub>O measurements using an open chamber technique as described by Menéndez *et al.* (2006) using a chemi-luminescence analyser (AC31M Environment S.A) during 15 days.

Ammonia emissions started to be measured one hour after treatment applications using an open chamber technique. Measurements took place for 44 hours. Concentrations of NH<sub>3</sub> were measured at the air inlet and outlet of the chamber using a photoacoustic infrared gas analyser (Brüel and Kjaer 1302 Multi-Gas Monitor).

Cumulative gas emissions for all the gases were estimated by averaging the rate of loss between two successive determinations, multiplying that average rate by the length of the period between the measurements, and adding that amount to the previous cumulative total. Differences between cumulative gas emissions in the different treatments were compared by ANOVA as well as Duncan tests for separation of means between treatments ( $P < 0.05$ ).

## Results and discussion

Untreated slurry showed the highest ammonia volatilization rates immediately after fertilization. Cumulative emissions were 63% and 80% lower with the application of the solid and the liquid phases, respectively (Table 2). The lower NH<sub>3</sub> emission rates observed by the application of the electroflotation products, with respect to the original untreated slurry, were probably due to the physical characteristics of these products. As reported by many authors (Moal *et al.*, 1995; Braschkat *et al.*, 1997; Vandr  and Kaupenjohann, 1998) a higher dry matter content in the slurry would make its infiltration into the soil more difficult, enhancing NH<sub>3</sub> volatilization losses. In this context, de Jonge *et al.* (2004) suggested that a greater infiltration of the slurry into the soil could decrease NH<sub>3</sub> volatilization. Furthermore, they were similar to those of the control treatment, which indicates a great efficiency in decreasing nitrogen losses by NH<sub>3</sub> volatilization.

Cumulative N<sub>2</sub>O emissions were shown to be higher after the application of the different treatments with respect to the control (Table 2), but no significant differences were observed between the untreated slurry and the products of electroflotation. Higher N<sub>2</sub>O losses by denitrification could have been expected in the untreated slurry because of its higher dry matter content. In contrast, the untreated slurry induced even slightly, although not significantly, lower losses than the solid phase. This was possibly due to the fact that in the solid phase, as well as in the liquid, the carbon present was presumably easily available carbon for the denitrifying organisms, which may have enhanced denitrification to the same level as in the untreated slurry.

Because of the low NO emission rates measured during this study, no differences were observed between treatments. Cumulative losses over the 15 days duration of the assay, therefore, did not show any statistically significant differences between treatments. Although the products of electroflotation induced slightly higher NO losses than the untreated slurry, they were not statistically different even from the control, which showed the lowest losses (Table 2).

Table 2. Cumulative emissions of the different gases in spring (Experiment 1) up to day 15 after application of treatments, and CO<sub>2</sub> equivalents (CO<sub>2</sub>e) for cumulative N<sub>2</sub>O emissions as well as total CO<sub>2</sub>e corresponding to N<sub>2</sub>O + CO<sub>2</sub> recommendations of IPCC (1996). Different letters within a column indicate significantly different rates ( $P < 0.05$ ;  $n = 4$ ).

Treatment	NH <sub>3</sub>	N <sub>2</sub> O		NO	CO <sub>2</sub>	Total CO <sub>2</sub> e Mg CO <sub>2</sub> e ha <sup>-1</sup>
	44 hours kg N ha <sup>-1</sup>	15 days kg N ha <sup>-1</sup>	CO <sub>2</sub> e Mg CO <sub>2</sub> e ha <sup>-1</sup>	15 days g N ha <sup>-1</sup>	15 days Mg ha <sup>-1</sup>	
Control	0.30 b	1.20 b	0.58	45.30 a	4.11 b	4.69 b
Untreated slurry	1.17 a	2.50 a	1.22	62.48 a	4.99 a	6.21 a
Solid phase	0.43 b	2.89 a	1.41	77.13 a	4.84 ab	6.25 a
Liquid phase	0.24 b	2.36 a	1.15	73.86 a	4.58 ab	5.73 a

When cumulative CO<sub>2</sub> emissions were calculated (Table 2), the untreated slurry was shown to have induced significantly higher losses than the control. The products of electroflotation showed lower emissions than the untreated slurry, although these differences were not significant. When the global warming potential was calculated, expressed in terms of CO<sub>2</sub> equivalents corresponding to the global effect induced by both CO<sub>2</sub> and N<sub>2</sub>O together, no significant difference was observed between the products of electroflotation with respect to the untreated slurry.

## Conclusions

The solid and liquid phases of electroflotation can be considered useful products as fertilizers. In the short term both products do not increase the risk of global warming through increased greenhouse gas (N<sub>2</sub>O, NO and CO<sub>2</sub>) emissions. The emissions induced by their application are of the same magnitude as those induced by the application of the original slurry. By contrast, their application has a positive environmental effect in terms of reduced NH<sub>3</sub> emissions.

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## Effects of direct seeding and cow slurry application on maize production in fine-textured soils

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### Abstract

In many areas of the world, it has been shown that the utilization of direct seeding (no-tillage) and organic fertilization (animal waste as fertilizer) in agroecosystems can reduce costs and soil organic carbon losses. The aim of this study was to evaluate the effects of direct seeding and cow slurry utilization on forage maize yield and soil quality in fine-textured soils under a temperate and humid climate. In order to do this, a 3-year field assay, that consisted of an intensive crop rotation (cereals in winter/forage maize in summer), was established in Derio (Basque Country). The effects of these practices on forage maize production and soil compaction were compared throughout the experimental period with those induced by mineral fertilization and conventional tillage. Under no-tillage, a compacted top layer was observed from the first year. During the first two years, this compaction did not have any impact on forage maize production; however, in the third year, forage maize production was significantly reduced under no-tillage (53% and 67% in organic and mineral plots, respectively). Fertilization with cow slurry resulted in values of forage yield similar to those obtained with mineral fertilization.

Keywords: maize, direct seeding, slurry, soil compaction, tillage

### Introduction

It is a well-known fact that organic fertilization and no-tillage can have beneficial effects on soil functioning. In this respect, the utilization of animal slurry to fertilize soils, apart from helping to reuse a waste, is known to increase the content of soil organic matter (OM) as well as the microbial biomass and activity. In turn, no-tillage, apart from saving fuel and labour costs, has many other advantages such as controlling erosion and soil moisture loss, and increasing soil OM and aggregate stability. However, to our knowledge, no-tillage practice is not widely used in areas with fine-textured soils (susceptible to soil superficial compaction) such as those characteristic of the Basque Country (northern Spain). In compacted soils, plant root length is reduced and, as a consequence, the roots explore a relatively small soil volume and then intercept a limited amount of water and nutrients. Furthermore, soil compaction can also decrease microbial biomass and enzyme activity in the soil. As a consequence, mineralization is depressed and nutrient uptake by the growing crop is further reduced, finally resulting in poor crop yields. The main objective of the current work was to study the effects of the utilization of cow slurry and no-tillage on forage maize production and soil quality in fine-textured and humid soils.

### Materials and methods

In spring 2005, a 3-year field assay, located in Derio (Basque Country), was established in a former grassland that had been seeded with Italian ryegrass (*Lolium multiflorum* Lam. cv. Nival) without any fertilization. The soil showed the following initial characteristics: silty clay loam texture (33% clay), pH = 5.1 (1:2.5 in water), 17.2 g OM kg<sup>-1</sup> DM soil, 1.3 g total N kg<sup>-1</sup> DM soil, 8.5 mg Olsen P kg<sup>-1</sup> DM soil, and 125 mg extractable K (NH<sub>4</sub>Ac) kg<sup>-1</sup> DM

soil. The field assay consisted of an intensive crop rotation (cereals in winter/forage maize in summer). The following treatments were applied: (i) conventional tillage + mineral fertilization, (ii) conventional tillage + cow slurry, (iii) no-tillage + mineral fertilization, and (iv) no-tillage + cow slurry. A randomized complete block design (5 x 3.5 m experimental plots) with 3 replicates was used for this experiment. Prior to the establishment of each crop, a mineral fertilization treatment, consisting of the addition of 150 kg N ha<sup>-1</sup>, 100 kg P ha<sup>-1</sup>, and 150 kg K ha<sup>-1</sup>, was applied. Similar doses were applied in the organic fertilization treatment with fresh cow slurry (considering the ammonium N-content). For the conventional tillage, soil was ploughed to 25 cm. Direct sowing for the no-tillage treatment was carried out with a Semeato machine. Pre-emergence herbicide application consisted of 8 L ha<sup>-1</sup> of 36% (w/v) glyphosate. During the growing period of maize (*Zea mays* L. cv. Nival), soil sampling was carried out one and a half months after tillage and fertilization treatments throughout the experimental period (three years). To this aim, 15 soil sub-samples were randomly collected from each plot, at a depth of 0-15 cm, and mixed together to get a composite sample for laboratory chemical analysis. Regarding soil physical parameters, five measurements of soil moisture and resistance to penetration (0-75 cm, with a Rimik CP40 penetrometer) were determined in each experimental plot. Penetration measurements were always carried out when the water content was at field capacity (3-5 days after a heavy rainfall). Finally, after harvest (3 months after treatment application), crop yield was determined. One-factor analysis of variance (ANOVA) and Fisher PLSD Post Hoc analysis were performed to establish significant differences ( $P < 0.05$ ).

## Results and discussion

Conventional tillage, as expected, provided a loose soil profile with penetration resistance values generally < 1.5 MPa for the 0-20 cm soil depth (Figure 1). By contrast, in those plots that had been subjected to direct seeding (no-tillage), a compacted top layer was observed from the very first year. Although penetrometers do not measure the exact pressure that plant roots must exert to penetrate the soil, they do provide a useful measure of the soil's resistance to root growth. In this respect, Boone and Veen (1982) found a close curvilinear relationship between maize root extension and cone resistance, and concluded that the lower critical limit of mechanical impedance for maize (the soil condition which causes a decrease in root growth rate of 50%) corresponds with a penetration resistance of 1.5 MPa. In this study, this threshold value of 1.5 MPa was exceeded in no-tillage plots from the very first year. Nevertheless, during the first two years, this compaction did not have any effect on maize yield (Figure 1) in agreement with other studies in which root growth was not hindered by soil strength at values lower than 2.5 MPa (Vepraskas, 1994). On the contrary, a considerable decrease in maize production was observed for the third year, when no-tillage plots reached values of soil penetration resistance close to 3.5 MPa. As a consequence, as far as crop production is concerned, these fine-textured soils would need to be subjected to tillage approximately every two years, in order to avoid their reaching high values of soil penetration resistance which cause severe restriction in root development. Most likely, the lower values of sand content (i.e., 14% sand in this study) are responsible for the soil compaction. After all, Dam *et al.* (2005), for a sandy loam soil in Canada, reported that long-term mean dry matter maize yields were not affected by tillage practices over an 11-year period. Similarly, during the first 5 years of their study, Linden *et al.* (2000) did not find any significant differences in crop yield between conventional tillage, reduced tillage and no-tillage plots. After the fifth year, they observed lower values of crop yield in no-tillage plots corresponding to a silt loam soil in Minnesota. In our study, within each tillage system, significant differences in soil compaction and maize yield were not observed between mineral and organic fertilization treatments. During the third year, in which soil compaction critically hindered root develop-

ment, the application of cow slurry seemed to alleviate this problem, as maize yield was approximately 40% higher in no-tillage plots fertilized with cow slurry as compared to no-tillage plots subjected to mineral fertilization. Furthermore, the addition of cow slurry increased soil pH and the content of soil OM (data not shown). Jiang-Tao *et al.* (2007) also found that application of organic manure could improve the soil's stability and mechanical properties.

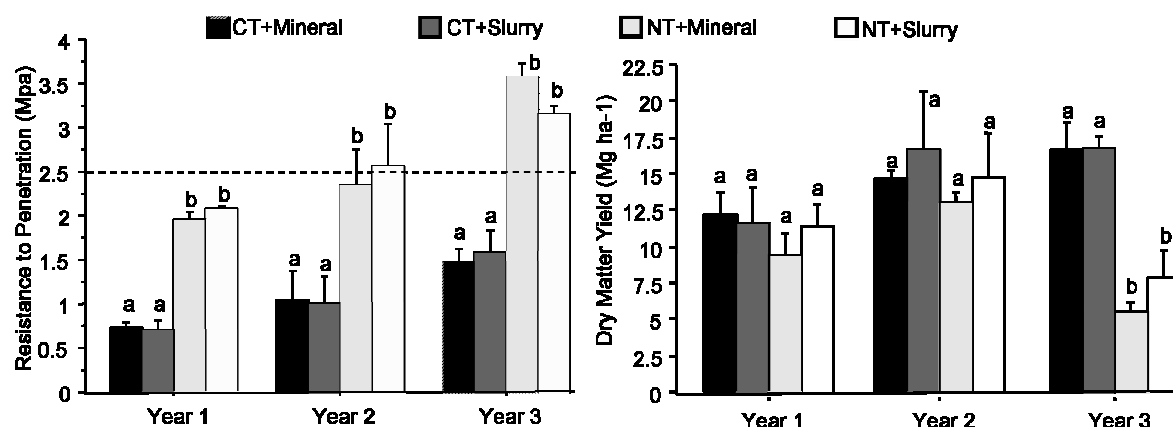


Figure 1. Effects of tillage and fertilization treatments on soil resistance to penetration and maize yield. Bars with different letters in each year indicate significant differences ( $P < 0.05$ ). CT: conventional tillage; NT: no-tillage.

## Conclusions

No-tillage increased soil compaction from the very first year as compared to conventional tillage. During the first two years, this compaction did not have any negative effect on maize yield. By contrast, forage maize production was significantly reduced in the third year. Under no-tillage, compared with mineral fertilization, the addition of cow slurry could lead to higher values of forage crop production and improves soil physicochemical properties.

## Acknowledgements

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## Algae residue to enhance pasture production

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### Abstract

An experiment is described which aims to evaluate the effects of different rates of algae residue, combined with different mineral fertilizer rates, and to deliver best combinations for improved pasture production. It was found that mineral fertilizer should be used with the algae residue to promote pasture production if a low dose of residue is used. The composition and its degree of mineralization of algal sludge should be evaluated before using it to promote crop production.

Keywords: acid soils, pH

### Introduction

EU regulations to improve continental water quality have made it compulsory to clean the water used by industries before discharging it. The residue generated can be used in grasslands to improve pasture production. Due to the different kind of industries, residues can have different characteristics due to their origin, but also due to the product delivered by the industry. For example, dairy industries have high nitrogen content which has been successfully evaluated in pasture production. Algae processing industries produce a different kind of residue, which has low nitrogen content, but which can be used successfully in grasslands developed on acid soils due to its high pH. CEAMSA is an industry situated in NW Spain which manufactures and supplies a comprehensive range of high quality carrageenan products, speciality hydrocolloid blends and refined locust bean gum to the global food industry. Main resource of carrageenan products are algae. During the processing of carrageenan products a residue is obtained, which has high pH and could be recycled in agriculture. Most of NW Spain soils are characterized by having a very low pH, due to the high level of precipitation in the area. The use of high pH residues could, therefore, be used to restore soil fertility capacity.

The aim of this experiment was to evaluate over two years the effect of the use of an industrial algae residue on pasture production.

### Materials and methods

The experiment was established in November 2004 in Pastoriza county (Lugo, Spain) and located in the Atlantic biogeographic region. The initial soil pH was 5.5. The experimental design was a randomized block with 10 treatments and four replicas. The main objective was to evaluate the effects of 5 rates of algae sludge (0, 4, 8, 12, 16 and 20 t ha<sup>-1</sup>) applied in autumn 2004 and winter 2005, either alone or combined with mineral fertilization (thus 10 treatments in total), on pasture production during two years. Mineral fertilization consisted of an application of 500 kg ha<sup>-1</sup> of 8:24:16 (N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O) fertilizer in February, plus 40 kg N ha<sup>-1</sup> after each spring harvest with the exception of the last spring harvest. No fertilization was applied in autumn. Two treatment controls were also established: a) lime application (traditionally used in the area) plus mineral fertilization, and b) lime application without mineral fertilization. The algae residue, as well as the lime application, was made before the traditional sowing in autumn 2004. The algae residue had a pH (water) of 9, a dry

matter content of 24.65% and reduced levels of N (0.15%), K (0.26%), and P (0.02%) when compared with municipal sewage sludge. However, Na levels were higher than for municipal sewage sludge (0.15%), probably due to the marine provenance of the algae. Ca and Mg concentrations were 0.6% and 1.7%, respectively. The main pasture sowing mixture was 25 kg ha<sup>-1</sup> of perennial ryegrass (*Lolium perenne* L.) and 4 kg ha<sup>-1</sup> of white clover (*Trifolium repens* L.). Three pasture samplings were made during 2005 and 2006, which consisted of harvesting an area of 4 m<sup>2</sup> in each plot twice in spring and once in autumn. Cut pasture was weighed in the field and samples were taken to the laboratory to determine dry matter content and botanical composition. Soil samples were taken at the end of 2006 to a depth of 25 cm, as required by Spanish regulations for the use of residues in agriculture (RD 1310/90). Aluminium saturation percentage was estimated in BaCl following the method of Guitian and Carballas (1976). ANOVA and Duncan tests were performed for statistical analyses.

## Results and discussion

The aluminium percentage-saturation is shown in Figure 1. Its low value (below 30) indicates that this variable should not limit pasture production. Medium and high doses of the algae residue reduced the aluminium saturation percentage. This could be explained by the high pH of the residue. At low doses, mineral fertilization increased the percentage of aluminium, which could be explained by the extractions, having been derived from pasture production, were not compensated by the low algae dose applied.

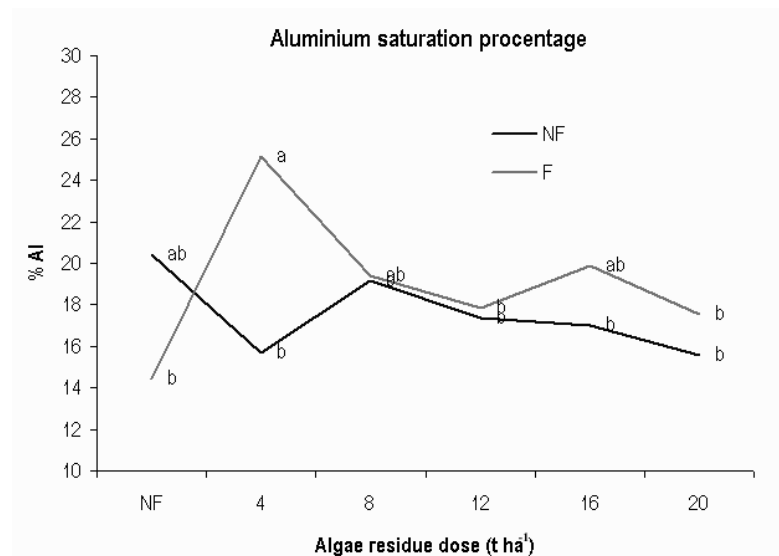


Figure 1. Aluminium saturation percentage after two years of the experiment and for the different treatments (inputs of 4, 8, 12, 16 and 20 t ha<sup>-1</sup> algae residue, complemented (top line, F) or not (NF) with mineral fertilizer (8N:24P<sub>2</sub>O<sub>5</sub>:16K<sub>2</sub>O). Different letters indicate means significantly differences between treatments ( $P < 0.05$ ).

Pasture production in the two years of study can be seen in Figure 2. Pasture production was only modified by the fertilization input during the first harvest due to the lack of residual effect of the mineral fertilizer (Mosquera-Losada *et al.*, 1999). This effect was found in both years of the study. However, pasture production was enhanced under the medium dose (12 t ha<sup>-1</sup>) of algae sewage sludge in the first year. This could be explained by the lack of incorporation of the sludge at the high rates of 16 and 20 t ha<sup>-1</sup>, which physically limited pasture production in the two years of study. Mineral fertilization enhanced pasture production in all the treatments up to 16 t ha<sup>-1</sup> during the second year. This could be explained by the combination of the residual effect of the algae when applied in consecutive years.

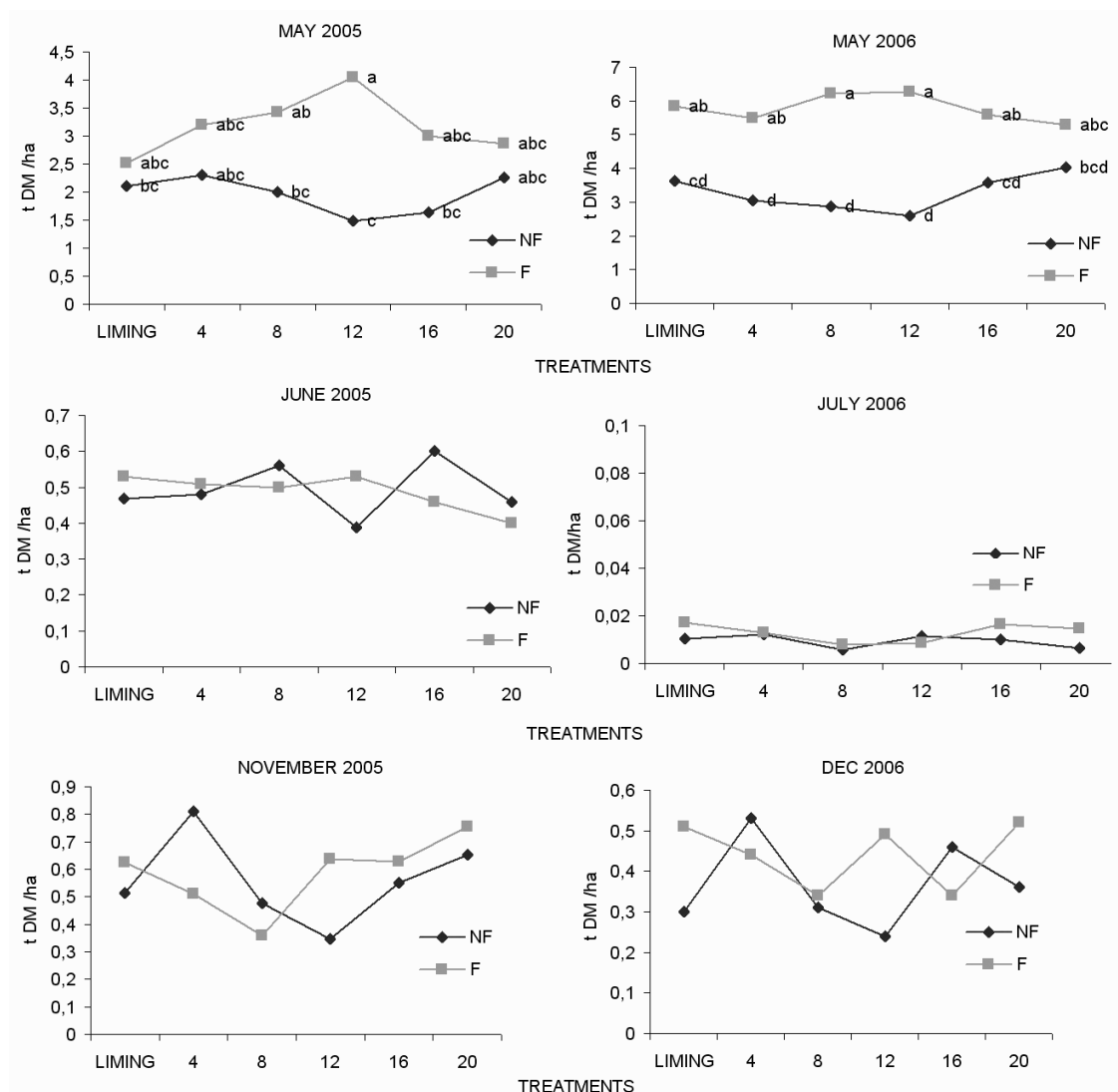


Figure 2. Pasture production in the different harvests of 2005-06 and in the different treatments (4, 8, 12, 16, 20 t ha<sup>-1</sup> algae residue), complemented (F) or not (NF) with mineral fertilizer (8N:24P<sub>2</sub>O<sub>5</sub>:16K<sub>2</sub>O). Different letters indicate significant differences between treatments ( $P < 0.05$ ).

## Conclusions

Inputs of algae residue help to maintain soil fertility, as found in the percentage saturation of aluminium. However, residues with lime capacity should be tested at a medium and long term in order to determine their benefits from a pasture production point of view.

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# **Productive and economic results for organic farms with a large share of grassland in Poland in the years 2004-2006**

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## **Abstract**

Questionnaire studies were carried out on 34 organic farms in the years 2004-2006. Mean proportion of grasslands in total croplands was 49.3% (Poland average is 21%). Productive and economic results of these farms are presented against a background of natural and agricultural characteristics. Studies were aimed at assessing meadow organic farms against traditional, non-organic farms. Standard gross margin was adopted as the main criterion of economic evaluation for the year 2005. The standard gross margin for this period, calculated per ha of croplands and per person permanently employed in a farm, showed an increasing trend. With the enlargement of farm area the former index decreased while the latter increased. In general, our studies showed that satisfactory economic results could be obtained in most studied farms providing state subvention and subsidies from the European Union.

Keywords: effectiveness of capital assets, grasslands, organic farming, Standard Gross Margin

## **Introduction**

Legally guaranteed subsidies to organic farming and an increasing market for healthy food after Polish accession to the European Union were the reasons for increasing the number and area of organic farms, and there are chances for their further development. Already functioning organic farms with animal production in Poland have markedly greater share of permanent grasslands than traditional (non-organic) farms. It is beneficial since animal production is an integral part of organic farming. Animal production may be directed to cattle for slaughter cattle (low input systems) or to milk production for dairy products of high quality (Jankowska-Huflejt, 2007). Ecological food produced in Poland may be competitive in both quality and price. The main hypothesis was that organic farms are able to produce ecological food and to achieve satisfactory economic results comparable to those for traditional farms.

## **Materials and methods**

Studies were carried out with the questionnaire method in the years 2004-2006 on 34 organic farms in the whole country (Zastawny and Jankowska-Huflejt, 2005; Jankowska-Huflejt, 2006, 2007). Farms with animal production based on their own grasslands which covered no less than 30% of croplands were selected for this study. Farm area varied from 3.13 ha to 319.42 ha (mean 35.27 ha). Economic results of these farms are discussed against the background of their natural and agricultural characteristic. Standard gross margin (MAFF, 1992) for the year 2005 was adopted as the main criterion for economic assessment.

## **Results and discussion**

The most numerous of the studied organic farms were those having an area between 20.1 and 50.0 ha (mean 28.12 ha). Grasslands constituted nearly 50% of the total cropland area in these



farms (two times more than the average proportion in Poland). Crop structure was oriented to the production of bulk fodder for ruminants and own fodder grains, particularly oats, cereal mixtures, triticale and barley (Tables 1 and 2). Small farms (1-10 ha) were characterized by the greatest diversity of bred animals and by the largest animal stocking densities; the mean was 0.8 LU ha<sup>-1</sup> (LU = 500 kg live weight). In 7 farms the stocking rate exceeded 1 LU ha<sup>-1</sup> which is exceptionally high under conditions of Polish agriculture. The main livestock were dairy cattle of various breeds (Polish black-and-white, Polish red, Simmental, red-and-white, Holstein). Generally, the livestock on these farms were diverse as well as at a high stocking rate (2 to 2.5 times greater than the national average). Ruminants were the main kind of farm animals; therefore, providing them with good, balanced and healthy fodder was one of the most important factors for effective farming on organic farms. A low-to-medium level of investment in capital assets and relatively low level of direct costs for plant and animal production was found in the studied farms (Table 3). In spite of a large demand for labour, e.g. for pest control or plant protection, the farmers rarely employed hired workers which proves there was sufficient availability of labour resources in their own workforce.

Table 1. Cropland structure, crop structure (only relate to the use of arable lands), animal stock and employment in studied organic farms in the year 2006.

Farm		Cropland structure, %			Crop structure, %			Animal stock LU/ha croplands	Employment per100 ha
Area, ha	No.	Arable lands	Grass- lands	Orchards and gardens	Cere- als	Fodder crops	Root crops		
1.1-10.0	7	41.0	56.5	2.5	41.4	41.4	12.0	0.80	38.9
10.1-20.0	9	57.1	40.7	2.2	33.8	31.7	11.6	0.65	16.1
20.1-50.0	11	34.8	64.7	0.9	55.8	33.8	6.6	0.67	6.8
> 50.0	7	58.4	41.6	0.0	49.7	39.7	0.9	0.66	2.6
Mean	34	47.8	50.9	1.4	45.2	37.3	3.5	0.70	15.00

Standard gross margin (mean of three years) for the studied farms, calculated per ha of croplands and per full-time employed person on the farm, was at a medium-to-low level. With increasing farm area this value decreased when calculated in terms of euro ha<sup>-1</sup> of croplands, but increased when calculated on a per-capita basis. Generally, incurred costs were not satisfactorily compensated by higher incomes from organic farming on all farms, but the efficiency of utilization of capital assets increased (Table 3).

Table 2. Standard gross margin (SGM) for 2005 for farms classified by farm area.

Group of farms, ha	Farm area, ha of croplands				Standard gross margin, euro/ha				
	2004	2005	2006	Mean	2004	2005	2006	Mean	STDV
1.0-10.0	6.99	6.43	7.30	6.91	291.0	1203.5	1130.8	875.0	507.1
10.1-20.0	14.34	13.00	12.12	13.15	404.3	939.5	715.8	686.5	268.8
20.1-50.0	26.04	28.49	29.84	28.12	360.8	602.0	502.5	488.5	121.2
> 50.0	78.78	120.84	110.21	103.28	149.5	431.5	287.3	289.5	141.0
Mean	29.88	38.86	37.06	35.27	287.0	741.3	644.0	546.5	239.2

## Conclusions

Among the studied organic farms (of low to medium investment in capital assets) the most numerous farms were those with an area of 20.1-50.0 ha (mean 28.12 ha) and having *ca.* 50% of their cropland area in grassland. Small farms (< 20 ha) were characterized by the most diverse stock and highest stocking rates and a prevalence of dairy cattle.

Table 3. Per capita standard gross margin and the effectiveness of capital assets (in euro of SGM per euro of capital assets) against farm area – years 2004-2006.

Group of farms, ha	Standard gross margin, euro/person					Capital assets, euro/ha	Effectiveness of capital assets				
	2004	2005	2006	Mean	Standard deviation		2004	2005	2006	mean	Standard deviation
1.0–10.0	871.5	3802.5	4455.0	3043.0	1908.7	6257.8	0.09	0.13	0.23	0.15	0.07
10.1–20.0	3433.3	8159.5	6132.5	5908.5	2371.1	4438.8	0.19	0.15	0.21	0.18	0.03
20.1–50.0	4931.3	9984.0	9727.5	8214.3	2846.0	1694.3	0.14	0.22	0.47	0.28	0.17
> 50.0	4386.3	9892.3	14799.3	9692.5	5209.4	1389.8	0.10	0.17	0.94	0.40	0.47
Mean	3524.5	7260.3	6824.3	5869.8	2042.7	3297.0	0.13	0.17	0.45	0.25	0.17

Standard gross margin was medium to low. Its value calculated per ha of croplands decreased with increasing farm area, but increased when calculated on the basis of per person employed on the farm. In the studied group only *ca.* 60% of farms conformed to the satisfactory economic requirement that per capita standard gross margin should exceed the 5,000 euro (Niewęłowska, 2005). These were usually small farms where economic barriers were a serious obstacle to realizing necessary investments. Economic results for these farms might increase with increasing incomes in the country's population, leading to increased purchasing of ecological products, and in western regions of Poland due to export potential to Western Europe.

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# The effects of red clover on winter wheat yields

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## Abstract

A trial was conducted on a light loamy dystic albeluvisol in western Lithuania to investigate the influence of a green manure (second cut of a sward based on 80% *Trifolium pratense* L. + 20% *Phleum pratense* L.) on the productivity of a following winter wheat crop in an organic farming system. The wheat crop in this organic system was compared with that in a 'sustainable farming system' in which mineral fertilizers and pesticides were supplied according to the plant needs, and both systems received 60 t ha<sup>-1</sup> of cattle manure. In both systems the swards were grown for hay, and independently of the farming system red clover content in the swards was high (98.0-99.4%). The dry matter yields of red clover regrowths after the first hay cut were low (1.17-1.24 t ha<sup>-1</sup>) because of low precipitation. A higher yield of winter wheat (6.72 t ha<sup>-1</sup>) was obtained in the sustainable farming system than the organic farming system (3.70 t ha<sup>-1</sup>). The maintenance of nutrients was better in the sustainable than in the organic farming system, where the lower wheat yield was attributed to the lower nutrient inputs; nitrogen, phosphorus and potassium, as supplied to the soil in the form green manure, were lower by 2.8, 6.1 and 1.1 times, respectively, in the organic system, compared with the nutrient inputs in the sustainable farming system.

Keywords: red clover, organic and sustainable farming systems, winter wheat

## Introduction

Humus reduction is often observed in intensive farming systems (Abdallahi *et al.*, 2000). In order to maintain soil fertility, farming systems that conserve organic material should be applied. Plants contribute to soil organic matter and, as a result, soil fertility depends on the residues of organic carbon from preceding crops and on its decomposition rate (Burke *et al.*, 1998). These processes are determined by different biological properties of plants, their chemical composition, soil chemical and physical properties and climate, as well as soil and crop cultivation management (Wivstad, 1999). Many authors suggest that perennial legumes, due to their abundant root residues rich in biologically fixed nitrogen, exert a positive effect on the soil lasting for several years. Therefore, to meet the high demand for nutrients it is rational to grow winter wheat after legumes. The second cut of a sward can be used as green manure for wheat. In an organic farming system the aftermath of a green manure-red clover crop was used as a source of fertilization for winter wheat and compared with a sustainable farming system in which mineral fertilizers were used. The objective of the present study was to determine value of red clover as a green manure for winter wheat grown on a dystic albeluvisol in western Lithuania.

## Materials and methods

Experiments were carried out at the Vezaiciai Branch of the Lithuanian Institute of Agriculture (region of West Lithuania) during 2001-2004. Red clover (*Trifolium pratense* L. cv. Liepsna) with timothy (*Phleum pratense* L. cv. Gintaras II), as an 80% red clover + 20% timothy mixture, were grown in the perennial grass phase of a rotation. Two farming systems were compared: 1) an organic farming system, without pesticides and mineral fertilizers, with 60 t ha<sup>-1</sup> solid cattle manure, in which the second cut was used as a green manure; and 2) a

sustainable farming system that also used the same rate of solid cattle manure (at 60 t ha<sup>-1</sup>) plus mineral fertilizers and pesticides according to the calculated plant needs. Treatments (farming system) were replicated 4 times. Mineral fertilizer needs were calculated according to soil agrochemical indicators: pH<sub>KCl</sub>, N<sub>total</sub>, humus, mobile P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O. The 'perennial grass phase' of the rotation preceded winter wheat, the rotation being: i) potatoes, ii) barley with undersown perennial grasses, iii) perennial grasses, and iv) winter wheat. The soil of the experimental site is a light loam (dystric albeluvisol). In the organic farming system, soil agrochemical characteristics were: pH<sub>KCl</sub> 6.3 ± 0.06, available P<sub>2</sub>O<sub>5</sub> 155 ± 5.60 mg kg<sup>-1</sup>, K<sub>2</sub>O 201 ± 11.4 mg kg<sup>-1</sup>, N<sub>total</sub> 0.11 ± 0.01%, humus 2.0 ± 0.12%. Soil agrochemical characteristics in the 'sustainable farming system' were: pH<sub>KCl</sub> 6.0 ± 0.07, available P<sub>2</sub>O<sub>5</sub> 191 ± 9.33 mg kg<sup>-1</sup>, K<sub>2</sub>O 201 ± 15.5 mg kg<sup>-1</sup>, N<sub>total</sub> 0.11 ± 0.01%, humus 2.0 ± 0.10%. Chemical composition of solid manure was: dry matter 28.9%, N 0.31%, P<sub>2</sub>O<sub>5</sub> 0.21%, K<sub>2</sub>O 1.16%. The weather conditions for the individual months of the growing season for the perennial grass phase of the rotation were diverse, with favourable conditions in April, May and June that were similar to the long-term average for temperature and rainfall. The most unfavourable conditions for herbage growth were in July, when the rainfall was by 50% lower, and the air temperature about 3 °C higher than the long-term average. The sward was cut twice. The first cut was taken in June, and the herbage was used as hay. The second cut was taken in July, at the beginning of the red clover flowering stage, and the herbage was used as green manure. Soil chemical analyses were determined annually after harvesting. Soil agrochemical properties (0-20 cm soil layer) were determined by the following methods: pH<sub>KCl</sub> – by electrometric method, available phosphorus and potassium by AL method, nitrogen by Kjeldhal, humus by Turin (ISO10390, 2001). Chemical composition of plants and farmyard manure were determined using the following techniques: nitrogen by Kjeldahl, phosphorus and potassium by flame photometry. The concentration of albumens in wheat grain was calculated by multiplying the nitrogen by the coefficient 5.70. The herbage dry matter (DM) yield was determined at each of the two cuts per year and the botanical composition was determined from each cut by hand-separating of herbage samples. The data on grain yield were adjusted to 14% moisture content.

## Results and discussion

Botanical sward analysis at each cut was carried out to estimate the effects of red clover as a preceding crop on winter wheat productivity in the two farming systems (Table 1). In both the organic and sustainable farming systems the content of clover in the swards at the first and second cuts was similar (range 98.0-99.4%).

Table 1. The effect of farming systems on red clover content, dry matter (DM) yield and metabolizable energy (ME).

Farming system	Cut of swards	Clover content in the sward (%)		DM yield t ha <sup>-1</sup>		ME GJ ha <sup>-1</sup>	
		$\bar{x} \pm S\bar{x}$	V (%)	$\bar{x} \pm S\bar{x}$	V (%)	$\bar{x} \pm S\bar{x}$	V (%)
Organic	1 <sup>st</sup> cut	98.2 ± 0.74	1.50	5.46 ± 0.27	9.80	44.6 ± 2.17	9.74
	2 <sup>nd</sup> cut	98.7 ± 0.53	1.08	1.17 ± 0.19	33.1	9.54 ± 1.58	33.0
Sustainable	1 <sup>st</sup> cut	98.0 ± 1.31	2.67	5.28 ± 0.31	11.8	43.1 ± 2.52	11.7
	2 <sup>nd</sup> cut	99.4 ± 0.51	1.02	1.24 ± 0.21	34.1	10.1 ± 1.73	34.2

$\bar{x}$  – Arithmetic mean,  $S\bar{x}$  – standard error the mean, V – coefficient of variation.

The yield of metabolizable energy (ME) for the separate plant yields was calculated for each cut in each farming system. The ME varied with the DM yield of perennial grasses and clover ( $y = 0.1224x + 0.0032$ ,  $R^2 = 0.990$ ,  $r = 0.995$ ). In both farming systems (organic and sustainable) the highest ME outputs (44.6 and 43.1 GJ ha<sup>-1</sup>) were harvested in the sward at the

first cut. The unfavourable weather conditions, particularly shortage of moisture, (rainfall of 48 mm was half of the long-term mean), resulted in a reduction in DM yield at the second cut of the sward, and the ME value of the harvested herbage also declined. The coefficient of variation of red clover content in the swards was identified for each cut in the two farming systems. Low coefficients of variation (1.02-2.67%) suggest that the proportion of red clover in the DM yield is stable. While analysing red clover DM yield and ME variation, it was noted that this was far greater than variation in clover content. In both the organic and sustainable farming systems the first cut of the swards was characterized by a medium variation in DM and ME content (variation coefficients 9.74-11.7%). However, for the second cut of both farming systems, the variation of DM and ME content was high (33.0-34.2%). When crops that have a high nitrogen requirement (potatoes, wheat) are grown in organic farming systems, there is a high chance that yields will significantly decrease. However, this may not happen if mineral fertilizer nitrogen is effectively replaced by farmyard manure or other organic fertilizers (Wivstad, 1999). In this experiment unfavourable weather conditions had a strong effect on the amount of green manure available from the aftermath of the red clover sward. Due to the shortage of rainfall, the herbage yield of the red clover aftermath in the organic farming system was low. The soil received 1.17 t ha<sup>-1</sup> of DM from the red clover crop, which corresponds to about 4 t ha<sup>-1</sup> of solid manure. Compared with the sustainable farming system in which mineral fertilizers were used, the soil in the organic farming system received from the clover green manure about 1.1 times less potassium, 6.1 times less phosphorus and 2.8 times less nitrogen. In the organic farming system the grain protein content was lower by 3.05 percentage points, and the winter wheat grain yield was by 1.8 times lower, than in the sustainable farming system.

Table 2. The effect of farming systems on the content of nutrients incorporated into the soil and on winter wheat yield and its quality.

Farming system	N	P (kg ha <sup>-1</sup> )	K	Wheat grain yield (t ha <sup>-1</sup> )	Concentration of albumens-N in wheat grain (as %)
Organic	31.8 ± 4.54	3.28 ± 0.40	27.4 ± 4.18	3.70 ± 0.14	9.35 ± 0.63
Sustainable	90.0 ± 1.50	20.0 ± 0.50	30.0 ± 1.60	6.72 ± 0.33	12.4 ± 0.68

## Conclusions

Red clover used as green manure did not have an acceptable effect on winter wheat yield, which in an organic farming system (3.70 t ha<sup>-1</sup>) was lower than the yield obtained in a sustainable farming system that used mineral fertilizers. The amount of red clover in the aftermath used for the green manure was low at 1.17 t DM ha<sup>-1</sup> due to low rainfall. This resulted in low quantities of nutrients being available for incorporation into the soil: N was 2.8 times lower, P was 6.1 times lower and K was 1.1 times lower than the amounts supplied in the sustainable farming treatment. The sustainable farming system provided a better nutrient supply from mineral fertilizers and this had a positive effect on grain yield of winter wheat, which amounted to 6.72 t ha<sup>-1</sup>.

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# The impact of legumes on cereals in a crop rotation and legume-cereal bi-cropping

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## Abstract

The potential of legumes to supply cereals with biological nitrogen was investigated in an experiment conducted at the Lithuanian Institute of Agriculture in Dotnuva. Barley, peas, white clover, red clover and perennial ryegrass were established in the first year of the experiment. In the autumn of the first year wheat was direct drilled into clovers and peas, forming a bi-cropping system, or was conventionally drilled after the preceding crops. In the spring of the third year, spring barley was direct or conventionally drilled into the plots. The grain yield of winter wheat depended on the preceding crop and sowing method. The best preceding crops for winter wheat were ploughed-in white and red clover. The highest amount of nitrogen in the grain yield was after ploughing in of clovers. The grain yield of winter wheat (575-1,445 kg ha<sup>-1</sup>) and spring barley (268-14,425 kg ha<sup>-1</sup>) was very low when they had been grown in the bi-cropping system; however, the N concentrations in winter wheat grain (18.2-19.3 g kg<sup>-1</sup>) and spring barley grain (21.2-23.3 g kg<sup>-1</sup>) were not lower than those in conventionally grown wheat grain (16.2-17.6 g kg<sup>-1</sup>) and barley grain (18.6-19.3 g kg<sup>-1</sup>).

Keywords: clovers, peas, cereals, bi-cropping

## Introduction

Legumes are one of the main plants for improving sustainability of agroecosystems. Legume swards are commonly sown with a cover crop grown for grain, and nitrogen (N) accumulated by legumes is utilized after they have been ploughed in, i.e. after 2-3 or more years (Kadziulis, 2001; Arlauskienė and Maikstenienė, 2006). Nowadays, under the conditions of sustainable or organic farming, it can be important to use the N accumulated by legumes as early and effectively as possible. Several experiments on direct drilling of cereals into growing white clover have been carried out in Europe with the aim of developing a bi-cropping system which allows reduced inputs of mineral fertilizers (Williams and Hayes, 1991; Clements *et al.*, 1995; Bergkvist, 2003; Thorsted *et al.*, 2006). As a result, legumes in a crop rotation would be a good alternative source of N to organic manures and mineral-N fertilizer, especially on farms where there are no sources of organic fertilizers. The aim of our experiment was to test the potential of annual and perennial legumes to supply cereal grain crops with biological N, without additional mineral N fertilization.

## Materials and methods

The experiment was conducted during 2004-2006 at the Lithuanian Institute of Agriculture in Dotnuva (55°24'N) on a loamy Endocalcari-Epihypogleyic Cambisol. Soil pH varied between 7.3 and 7.5, humus content was 2.3%, available P was 74-79 mg kg<sup>-1</sup> and K was 135-140 mg kg<sup>-1</sup>. Red clover (*Trifolium pratense* L.), white clover (*Trifolium repens* L.), perennial ryegrass (*Lolium perenne* L.), spring barley (*Hordeum vulgare* L.) and semi-leafless peas (*Pisum sativum* L.) were sown in the spring of 2004, as preceding crops before winter wheat (*Triticum aestivum* L.) (Table 1). The clovers were cut prior to under sowing of winter wheat, and the herbage was weighed and spread on the plots. In the autumn, winter wheat was sown

conventionally (CD) and direct drilled into legumes (DD), forming a bi-cropping system. Spring barley for grain was sown in 2006.

Table 1. Experimental design.

Wheat pre-crops, 2004	Winter wheat and sowing method, 2005	Spring barley and sowing method, 2006
Spring barley, B	Conventional drilling, CD	Conventional drilling, CD
Pea, P	Conventional drilling, CD	Conventional drilling, CD
Pea, P	Direct drilling, DD	Direct drilling, DD
White clover, Wcl	Conventional drilling, CD	Conventional drilling, CD
White clover, Wcl	Direct drilling, DD	Direct drilling, DD
Red clover, Rcl	Conventional drilling, CD	Conventional drilling, CD
Red clover, Rcl	Direct drilling, DD	Direct drilling, DD
Perennial ryegrass, Pr	Conventional drilling, CD	Conventional drilling, CD

The layout was a randomized block design with four replicates and a plot size of 12 m x 3 m. Dry matter (DM) yield was determined on the basis of total dry matter amount per plot and calculated as kg DM yield ha<sup>-1</sup>. The inorganic nitrogen in the DM yield was determined by the Kjeldahl method. The experimental data were statistically processed using ANOVA.

## Results and discussion

Winter wheat grain yield was significantly higher after ploughed-in white and red clovers than after barley. Winter wheat grain yield was on average 294 kg ha<sup>-1</sup> higher after peas as the preceding crop, than after barley (Table 2).

Table 2. The effect of white and red clover on winter wheat and spring barley grain yield.

Wheat pre-crops <sup>1)</sup> + sowing method <sup>2)</sup>	Winter wheat, 2005			Spring barley, 2006		
	Grain yield kg ha <sup>-1</sup>	Nitrogen content in grain		Grain yield kg ha <sup>-1</sup>	Nitrogen content in grain	
		g kg <sup>-1</sup>	kg ha <sup>-1</sup>		g kg <sup>-1</sup>	kg ha <sup>-1</sup>
B – CD	2064	16.8	34.7	1560	19.1	29.8
P – CD	2358	16.9	39.8	1387	18.6	25.8
P – DD	575	19.3	11.1	268	23.3	6.24
Wcl – CD	3005	17.7	53.2	1647	18.9	30.5
Wcl – DD	1446	18.2	26.3	1102	22.2	24.5
Rcl – CD	3660	17.6	64.4	1781	19.3	34.4
Rcl – DD	948	18.4	17.4	1425	21.2	30.2
Pr – CD	1729	16.9	29.2	1540	19.2	29.6
<i>P</i> < 0.05	270.4	–*	4.75	284.3	–*	5.52

<sup>1)</sup>Pre-crops: B-barley, P-peas, Wcl-white clover, Rcl-red clover, Pr-perennial ryegrass.

<sup>2)</sup>Wheat sowing method: CD-conventional drilling, DD-direct drilling; –\* analyses were done on composite samples.

After having ploughed in the white and red clovers, wheat grain yield increased by 1,276 and 1,931 kg ha<sup>-1</sup>, respectively, compared with that after perennial ryegrass as the preceding crop. It was difficult to form a bi-cropping system when cereals had been sown into the growing first-year white or red clover; therefore, wheat yield in the bi-cropping system (clover-wheat) was poor. However, the results show that it is possible to achieve higher grain yield with white clover – wheat bi-cropping, than with red clover – wheat bi-cropping. Earlier findings noted that grain yield in a bi-cropping system is most often unstable and reaches only 50-60% of that when sown conventionally (Clements *et al.*, 1995; Kadziuliene and Kadziulis, 2006). To produce high grain yields, competition from the clover during tillering needs to be restricted to allow canopy expansion (Bergkvist, 2003).

Inclusion of legumes into crop rotations may be an effective means to significantly reduce mineral nitrogen use and increase nitrogen utilization efficiency (Neuens *et al.*, 2004). In our



experiment, in unfertilized cereal crops, when winter wheat had been grown after a preceding crop of clover in conventional sowing, or after direct drilled winter wheat, nitrogen concentration in the grain, in all cases, varied within the recommended range 18.2-18.4 g kg<sup>-1</sup>. However, after pea and grass as the preceding crops, nitrogen concentration was lower. A slightly higher nitrogen concentration in the grain yield was also determined when wheat had been grown with clover in a bi-cropping system. In research done in Denmark, when wheat and clover grew together, nitrogen concentration in grain was also higher by 0.11-0.39% than in the pure wheat crop (Thorsted *et al.*, 2006). A lower yield of nitrogen in grain was identified after having ploughed in a perennial grass crop (perennial ryegrass) compared with ploughing in of white or red clover crop. The least amount of nitrogen in grain per hectare was accumulated in the clovers-wheat bi-cropping system. Nitrogen amount in grain per hectare declined similarly to grain yield. Spring barley grain yield depended on the species of crops preceding winter wheat and, even more so, on the sowing method. In the experiment, barley grain yield of the conventional sowing was in the range 1,387 to 1,781 kg ha<sup>-1</sup>; however, when it had been grown in a bi-cropping system, the grain yield was somewhat lower. In the clover-barley bi-cropping system there was stiff competition from clovers; therefore the grain yield of clover-barley bi-cropping was significantly lower than that of conventionally sown barley after ploughed-in legumes. Having ploughed-in red clover and having sown barley conventionally, the grain yield was not significantly higher than that after ploughed-in perennial ryegrass.

## Conclusions

The winter wheat grain yield depended on the preceding crop and sowing method. The best preceding crops for winter wheat were ploughed-in white and red clover. The highest amount of nitrogen in the grain yield was recorded after ploughing-in of clovers.

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# Seasonality of productivity, botanical composition and N concentrations of four forage legume-grass mixtures under cutting

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## Abstract

Perennial ryegrass (*Lolium perenne*) was sown in pure stands and in 4 binary mixtures with birdsfoot trefoil (*Lotus corniculatus*), lucerne (*Medicago sativa*), red clover (*Trifolium pratense*) and white clover (*Trifolium repens*), respectively, in a randomized block experiment with three replicates on a clay soil in Wageningen in 2003. No N fertilizer was applied. The aim was to study seasonality of productivity, botanical composition and N concentrations in the various mixtures. Plots were cut four times in 2004 and five times in 2005. Annual DM yields of the mixtures ranged from 8.2 to 11.5 t ha<sup>-1</sup> in 2004 and from 8.2 to 16.3 t ha<sup>-1</sup> in 2005. The mixture with birdsfoot trefoil had the lowest yield, mixtures with red and white clover had the highest yield in 2004, and mixtures with red clover and lucerne had the highest yield in 2005. In all mixtures, the legume proportion increased in successive cuts but declined in the final cut in October. The N concentration in grass was always lower than in any legume. The N concentration in grass from mixtures was positively related to the proportion of legume, and to total yield. Grass N concentration increased during the season, both in pure and mixed stands. This study shows the potential of such mixtures in cutting systems under a low N input management. Birdsfoot trefoil was not persistent in this experiment.

Keywords: clover, lucerne, birdsfoot trefoil, N, species composition, seasonal yield pattern

## Introduction

In the search for quality legume-based forage systems in contrasting environments, an experiment was established in 2003 with one grass and four legume species. The aim was to study seasonality of productivity, botanical composition and N concentrations in the various mixtures under a cutting regime without N application.

## Materials and methods

Pure perennial ryegrass (*Lolium perenne*; Lp; cv. Fennema) and binary mixtures with *Lotus corniculatus* (Lc; cv. Rocco), *Medicago sativa* (Ms; cv. Daisy), *Trifolium pratense* (Tp; cv. Pirat) and *Trifolium repens* (Tr; cv. Klondyke) were sown in 3 replicates on a clay soil in Wageningen, The Netherlands, in September 2003. The trial was part of EU COST-action 852, WG3. Plot size was 3 by 8 meters. No N fertilizer was applied. P and K were applied according to requirements. The plots were cut 4 times in 2004 and 5 times in 2005 and 2006 with a forage plot harvester (Haldrup, Løgstør, Denmark). Pluck samples were collected to assess botanical composition; for each plot a subsample of 250 g was hand-separated into Lp, the sown legume spp. and unsown species. Another sample of 250 g was taken with a grass core to assess herbage DM percentage. All samples were oven-dried at 70 °C for 48 h. The hand-sorted pure grass and legume samples of the three last cuts of 2004 and all cuts of 2005 and 2006 were ground over a 1 mm mill (Peppink, Deventer, the Netherlands) and analysed with NIRS for N concentration. Results were analysed for each year as a replicated random block design with legume treatment as main factor.

## Results and discussion

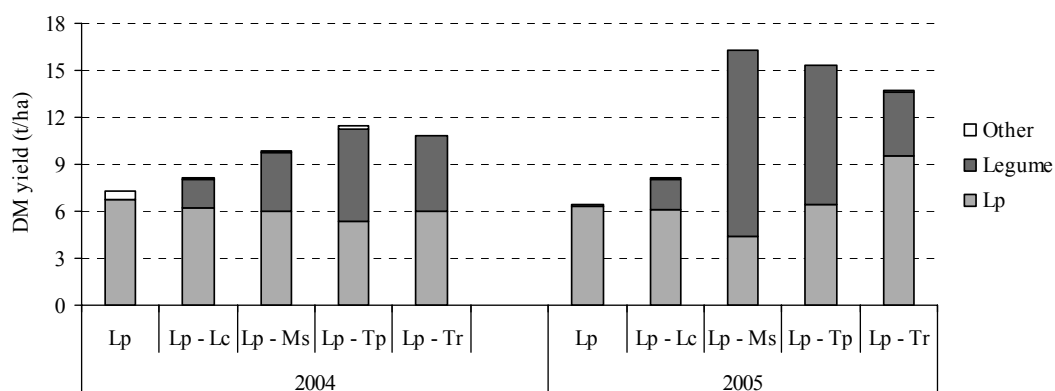


Figure 1. Yields of *Lolium perenne* (Lp) and binary mixtures with *Lotus corniculatus* (Lc), *Medicago sativa* (Ms), *Trifolium pratense* (Tp) and *Trifolium repens* (Tr) in 2004 and 2005.

Pure perennial ryegrass always yielded least, although in both years Lp-Lc was not different ( $P > 0.05$ ) from Lp (Figure 1). Lp-Tp and Lp-Tr were the highest yielding mixtures ( $P < 0.001$ ) in 2004, and Lp-Ms and Lp-Tp ( $P < 0.001$ ) in 2005. The productivity of mixtures with Ms, Tp and Tr increased after the first harvest year. Regarding component yield of mixtures, there was no difference in grass yield ( $P = 0.078$ ) in 2004. All mixtures produced *ca.* 6 t DM Lp ha<sup>-1</sup>. In 2005, the average grass yield was *ca.* 6.5 t ha<sup>-1</sup>, being highest ( $P < 0.001$ ) in mixtures with Tr and lowest with Ms. In both years, Lc yielded least (1.8 t ha<sup>-1</sup>) ( $P < 0.001$ ). In 2004, Tp yielded most (almost 6 t ha<sup>-1</sup>). In 2005, Ms yielded almost 12 t ha<sup>-1</sup>, and Tp was also high yielding (almost 9 t ha<sup>-1</sup>). Tr yielded around 4.4 t ha<sup>-1</sup> in both years.

The botanical composition of the plots at each cut during the two years is shown in Figure 2. In both years, the first cut had the highest ( $P < 0.001$ ) proportion of Lp. During the year the proportion of legume increased. Especially in mixtures with Tp and Ms this effect was very prominent. In 2005, these two species contributed  $< 50\%$  to the total yield in May, whereas they contributed  $> 90\%$  to the total yield in July. In the final harvest, the proportion of Lp generally increased except for Lp-Lc in 2004. The infestation with weeds was generally low, also because the plots were hand-weeded. Weeds never exceeded 10% of the total yield.

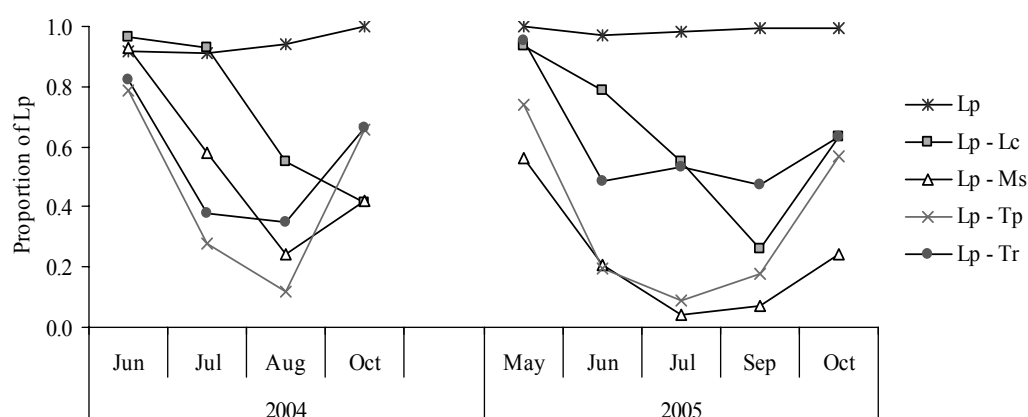


Figure 2. The proportion of *Lolium perenne* (Lp) in pure Lp stands and binary mixtures with *Lotus corniculatus* (Lc), *Medicago sativa* (Ms), *Trifolium pratense* (Tp) and *Trifolium repens* (Tr) in 2004 and 2005. Legume proportions (not shown) are complementary. Data are means of 3 reps.

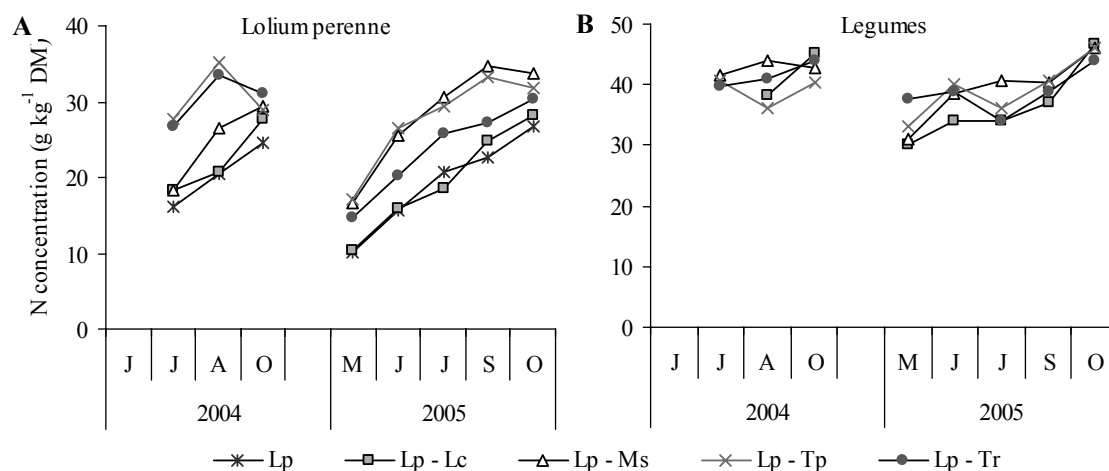


Figure 3A. Nitrogen concentration of *Lolium perenne* (Lp) in plots with pure Lp stands and binary mixtures with *Lotus corniculatus* (Lc), *Medicago sativa* (Ms), *Trifolium pratense* (Tp) and *Trifolium repens* (Tr) in 2004 and 2005. Figure 3B. N concentrations of the accompanying legumes in the same mixtures. Each point is the mean of 3 replicate plots.

The N concentration of Lp was generally lower than of the legumes (Figure 3). Lp had the lowest N concentration in pure stands; however the levels did not significantly differ from the Lp in mixture with Lc. The concentration of N in Lp was positively related ( $R > 0.95$ ) to the biomass produced by the accompanying legume within each year. The N concentration of the legumes (Figure 3B) was higher ( $P < 0.001$ ) in 2004 than in 2005. The N concentrations of the legumes did not differ to a great extent. The N concentration in Lp generally increased during the year (Figure 3A), both in mixtures and pure stands. Also the N concentration of the legumes showed a seasonal increase (Fig. 3B), although less pronounced than in Lp.

The results of this experiment are consistent with other cutting experiments (Heichel and Henjum, 1991; Elgersma and Hassink, 1997; Schils *et al.*, 1999) with grass-legume mixtures. Lc was not promising: the yields were low and the stands were vulnerable weed invasion. Tp and Ms yielded very well under the cutting regime applied in this study. For grazing situations (Brummer and Moore, 2000), white clover would probably be the most suitable species.

## Conclusions

In all mixtures the legume proportion increased during the year, except for the last cut in October. The N concentration in grass was generally lower than in legumes, and in mixtures it was positively related to the proportion of legume and to total yield. Herbage N concentrations increased during the season. This study shows the potential of mixtures of perennial ryegrass with lucerne, red, and white clover in cutting systems under a low N input management.

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# Evaluation of the contribution of soil organic matter mineralization to harvested N in temperate grasslands

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## Abstract

In temperate areas, grassland soils contain a large amount of organic N which contributes to sward nutrition if mineralized at the appropriate time. However, several field studies have shown that the amount of soil N available to swards fluctuates greatly from year to year. These variations are mainly a reflection of the great variability in organic matter mineralization rates which, for a given soil, depend mainly on temperature and soil moisture conditions, and the sward's N demand for growth. The objectives of this study were to monitor the net N fluxes associated with organic matter mineralization on two experimental plots laid out on permanent grasslands in the French Pyrenees and Massif Central, for 6 and 7 years respectively, to find out the extent to which the total amount of N provided by mineralization varies from year to year and to test whether temperature fluctuations can explain the variability in net N fluxes frequently observed. The results show that organic matter mineralization provides large amounts of N for sward growth at both sites. However, the amount varies from year to year. At both sites there is a significant relationship between the amount of N in plant biomass at the first cut and the average temperature in the previous two months, confirming that biological cycling in grassland soils is a temperature-controlled process. Large average temperature fluctuations may explain the big variability in forage production that occurs between years in unfertilized grasslands.

Keywords: grassland, nitrogen, organic matter mineralization, climate

## Introduction

Sustainable management of temperate grasslands relies on adequate utilization of nutrients present in soil reserves, since these pools can greatly contribute to herbage production under appropriate management practices. Besides organic manures and legumes, and in the absence of mineral fertilization, soil organic matter mineralization represents the main N source for sward nutrition (Whitehead, 2000).

Studies conducted *in situ* on a farm plot scale demonstrate that the soil's contribution to sward N nutrition can vary greatly between years, with large fluctuations in forage production as a consequence. Similarly, surveys carried out over several years on long-term field experiments show large year-to-year variability in the amounts of N provided from soil organic matter mineralization. Several hypotheses have been put forward to explain these results, including the effect of temperature on mineralization rates (Gill *et al.*, 1995). With this in mind, our objectives were to determine the net N fluxes associated with organic matter mineralization from a measurement of N in plant biomass, to verify the extent to which the total amount of N provided by mineralization varies between years, and to test whether temperature fluctuations can explain the variability in net N fluxes frequently observed.

## Materials and methods

The study used two experimental plots laid out on permanent grassland. One grassland experiment has been located since 1999 close to the village of Ercé in the French Pyrenees (0°E, 43°N; elevation 660 m a.s.l.). The other, begun in 1998, is close to the village of

Gramond in the Massif Central, France (2°E, 44°N; elevation 607 m a.s.l.). The mean annual air temperatures are 12.7 °C and 11.0 °C, and the mean annual rainfall is 1,200 mm and 960 mm at the Ercé and Gramond sites, respectively. Before the experiments started, neither site had received mineral fertilization in the previous 10 years, although at the Ercé site the soil had received farmyard manure every year. At Ercé, the soil was a Luvisol developed on alluvium, and at Gramond a Brunisol developed on mica schist. These soils exhibited features characteristic of permanent grasslands, with high organic matter and N contents: 95.2 mg g<sup>-1</sup> and 5.5 mg g<sup>-1</sup> respectively for Ercé, and 63.5 mg g<sup>-1</sup> and 3.3 mg g<sup>-1</sup> for Gramond (0-5 cm). Both soils are deep and well-drained, with a topsoil of silty loam at Ercé and sandy clay loam at Gramond. Herbage production was monitored under unfertilized conditions as the experiment was intended to study the capacity of soil to supply N to the sward over time with frequent defoliation (4 times a year) and in two contrasting N regimes (negative and balanced). The negative budget (NB) treatment is achieved when all the biomass production is exported from the plots, whereas a balanced budget (BB) treatment corresponds to plots in which all the biomass produced is returned to the plot immediately after harvest as mulch. The plots were arranged in four randomized blocks. Before each harvest, dry matter (DM) yield was measured on each plot by cutting the sward within a 0.25m x 0.75m quadrat with edging shears; herbage was oven dried for 48 h at 80 °C, ground to 0.5 mm and then analysed for N content (%) with a CN gas analyser (LECO Corporation, St Joseph, Michigan, USA). The N off-take for an individual cut was calculated by multiplying the dry matter yield by its N content. Total annual N off-take is obtained from the sum of the N off-takes for each individual cut.

## Results and Discussion

The total annual amount of N removed in forage is presented in Figure 1 for each year from the beginning of the experiments. The data confirm that the amount provided is large and variable from year to year. For both sites and for a given year, the total amount of N in plant biomass measured for the NB treatments was always less than that obtained for BB, except at Ercé in 1999 and 2000. At Ercé the maximum amount was observed in 2001 (285 ± 41 kg N ha<sup>-1</sup>) for the BB treatment, and the minimum (110 ± 40 kg N ha<sup>-1</sup>) in 2004 for NB. At Gramond, N in plant biomass varied between a maximum of 318 ± 61 kg N ha<sup>-1</sup> in 1999 for BB plots and a minimum of 60 ± 15 kg N ha<sup>-1</sup> measured in 2003 for the NB treatment. A decreasing trend for total annual plant N was noticed over time for both treatments, although it was more accentuated at Gramond. Such behaviour, expected for the NB treatment where the amount of potentially mineralizable N decreases with time, is related to severe heat and moisture stresses, which greatly affected forage production in 2003 and 2004 at both sites.

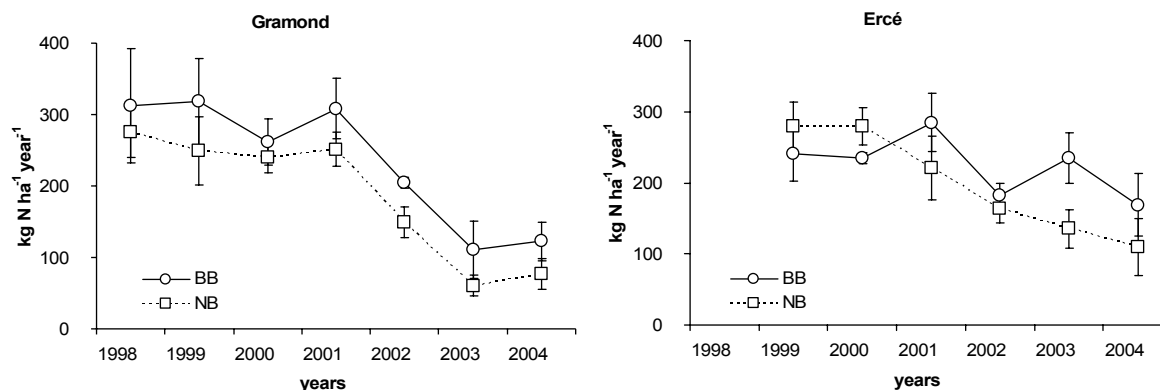


Figure 1. Average total annual amount of N in plant biomass and standard deviation (kg ha<sup>-1</sup> year<sup>-1</sup>; n = 4).

In order to ascertain the extent to which year-to-year variability was due to fluctuations in average temperature between years, we plotted the amount of N in plant biomass at the first cut for the BB treatment as a function of the average temperature measured over the previous 60 days; this cycle being the most appropriate for that purpose, since it represents 32% and 42% of the total annual biomass production at Ercé and Gramond, respectively. At Grammond, forage production measured at the first cut varied between 5.6 t ha<sup>-1</sup> (2001) and 2.5 t ha<sup>-1</sup> (2002), whereas at Ercé, it fluctuated between 7.5 t ha<sup>-1</sup> (2003) and 2.4 t ha<sup>-1</sup> (2000). For both fields, the first growth cycle represents the period when water stress is less likely to occur. However, for Gramond, data obtained in 2002 and 2003 are not included in the relation since water stress occurred during that cycle. Results presented in Figure 2 show for both sites that N removed in forage increases when the average air temperature in the 60 days preceding the cut increases, suggesting that the N provided to the sward from organic matter mineralization increases with temperature. The temperature range is similar for both sites; however, we observe that the slope of the relation is different: one degree increase in the average temperature increases the N budget by 12 kg at Ercé and by 22 kg at Gramond. One possible explanation for this difference is the chemical nature of the mulch, which contained legumes (3 years out of 7) at Gramond.

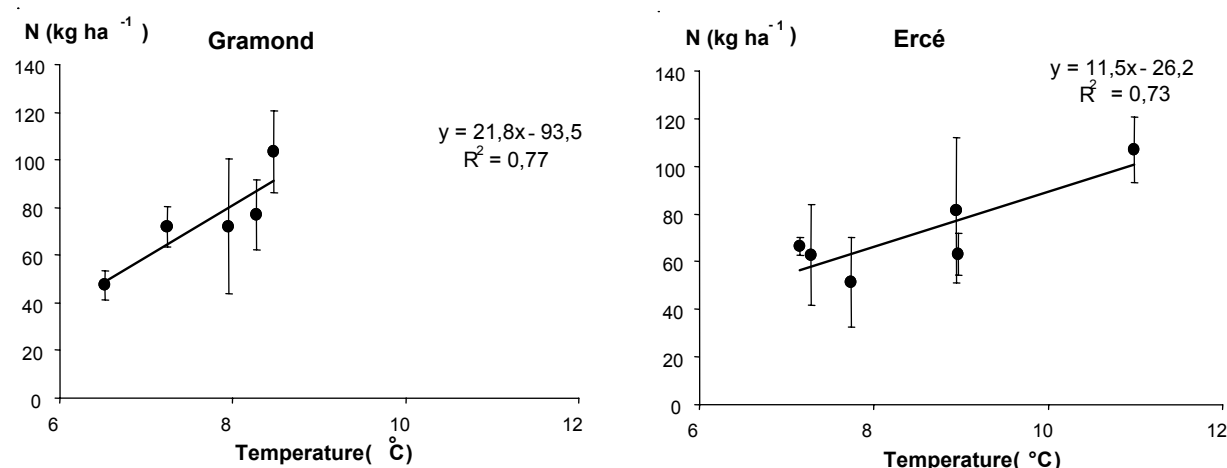


Figure 2. Relation between the average amount of N in plant biomass at the first cut and the average temperature measured over the previous 60 days for balance budget treatment plots (BB).

## Conclusions

Surveys carried out on experimental plots laid out on permanent grasslands demonstrate that organic matter mineralization can supply very large amounts of N to swards. For both sites there was a significant relationship between the amount of N removed in forage at the first cut and the average temperature in the previous two months, confirming that biological cycling in grassland soils is a temperature-controlled process. Large average temperature fluctuations may explain the big variability in early forage production (in the first growth cycle) between years in unfertilized grasslands.

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## Inter-annual variations in phosphorus content of semiarid grasslands over a long time period

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### Abstract

Mediterranean grasslands of the 'dehesa' ecosystem are characterized by strong annual and seasonal variations in biomass production and low fertility of the soil, particularly of phosphorus (P). Over a 19-year period (1987-2005), P content of 'dehesa' pastures in the province of Salamanca (western Spain) was analysed. Herbage samples were collected from several sites along a topographic gradient that differentiated two types of herbaceous communities on the upper and lower part of slopes. Phosphorus concentration in herbage ranged between 0.56-4.80 g kg<sup>-1</sup> DM in the upper zones, and between 0.56-3.50 g kg<sup>-1</sup> DM in the lower ones. The minimum mean P content was recorded in the lower zones in 1989 (1.42 g kg<sup>-1</sup> DM), and the maximum mean was recorded in the upper zone in 1988 (2.47 g kg<sup>-1</sup> DM). Inter-annual variations in P content in herbage at different altitude run parallel. Phosphorus content was significantly ( $P < 0.05$ ) correlated with the proportion of botanical groups in the pasture but not with any climatic variable related to precipitation along the sampling years. Phosphorus in herbage was correlated with soil characteristics in certain years.

Keywords: Phosphorus, long-term, Mediterranean pastures, dehesa

### Introduction

The 'dehesa' grasslands are characterized by the Mediterranean nature of the climate (dry summers and cold winters) and the low soil fertility, particularly of phosphorus (P). In these ecosystems, the pasture biomass production shows strong seasonal and inter-annual variations. Thus, variations in above-ground biomass production have been correlated with annual precipitation calculated from previous October to current June (Vázquez de Aldana *et al.*, 2006).

Phosphorus concentration in herbage varies with factors such as plant maturity stage, plant species, soil type and management (Whitehead, 2000). However, the information about inter-annual variations of mineral element contents in pastures is almost unknown. Following previous long-term studies of the 'dehesa' grasslands on nutritional quality (Vázquez de Aldana *et al.*, submitted), the objective of the present study was to analyse the variations in the P concentration of 'dehesa' pastures over a 19-year period (from 1987 to 2005) and to evaluate relationships with variables of climate, proportion of botanical groups in the pasture, and soil characteristics (available P, total carbon, organic matter, and coarse and fine sand fractions). These variations are important to understand functioning of pastures in the 'dehesa' ecosystem and for an adequate management.

### Materials and methods

Thirty gentle slopes were selected within the 'dehesa' grasslands of the province of Salamanca (western Spain). On each slope two topographically differentiated zones were determined: the uppermost and lowermost zones. These zones had different botanical composition: grasses dominated in the lower zones and forbs in the upper zones. The pastures were not fertilized.

Herbage samples were collected from 1987 to 2005 as follows: from 1987 to 1993 (1992 excluded) sampling was made in those thirty locations. From 1998 to 2005, five of the thirty previously selected slopes were selected and herbage samples were collected.

Plant samples were collected at the end of the growing season (late May or early June) at the time of the peak biomass. Sampling was accomplished by cutting the above-ground herbaceous vegetation in four randomly selected quadrats (0.25 m<sup>2</sup>). The samples were dried in a forced-air oven at 60 °C for 48 h and ground in a mill with a 0.5 mm mesh sieve. Phosphorus concentration was determined colorimetrically as molybdovanadate-phosphoric acid. In 1987, 1990 and 2005, soil samples were collected with cylinders 8 x 20 cm and analysed for available P, total carbon, organic matter, coarse and fine sand fractions.

The data were analysed statistically by analysis of variance for the effects of sampling year and slope zone. The relationship between P concentration and climatic variables and the proportion of grasses, legumes and forbs in the pasture were estimated using correlation analysis. The following climatic variables were considered: annual precipitation, seasonal and monthly precipitation, and number of days with precipitation (annual, seasonal and monthly). Annual precipitation was calculated in several ways: from January to December; from January to June (month of the harvest); and from previous October to current June. The relationships between P content in herbage and soil variables were statistically analysed for each year separately (1987, 1990 and 2005).

## Results and discussion

The P concentration varied in the communities of the upper and the lower zones along the sampling years (Figure 1). The analysis of variance showed a significant effect of slope position ( $P < 0.01$ ) and sampling year ( $P < 0.001$ ), and their interaction ( $P < 0.01$ ) on the P concentration. The mean P content ranged between 1.44 g kg<sup>-1</sup> (in 1990) and 2.47 g kg<sup>-1</sup> (in 1988) for the upper zones, and between 1.42 g kg<sup>-1</sup> (in 1989) and 2.06-2.07 g kg<sup>-1</sup> (in 2003 and 1993 respectively) for the lower zones. The overall mean was significantly ( $P < 0.01$ ) greater in the upper zone (1.98 g kg<sup>-1</sup>) than in the lower one (1.78 g kg<sup>-1</sup>), although as indicated by the ANOVA the differences between slope zones were only significant ( $P < 0.01$ ) in 1987, 1988 and 1989.

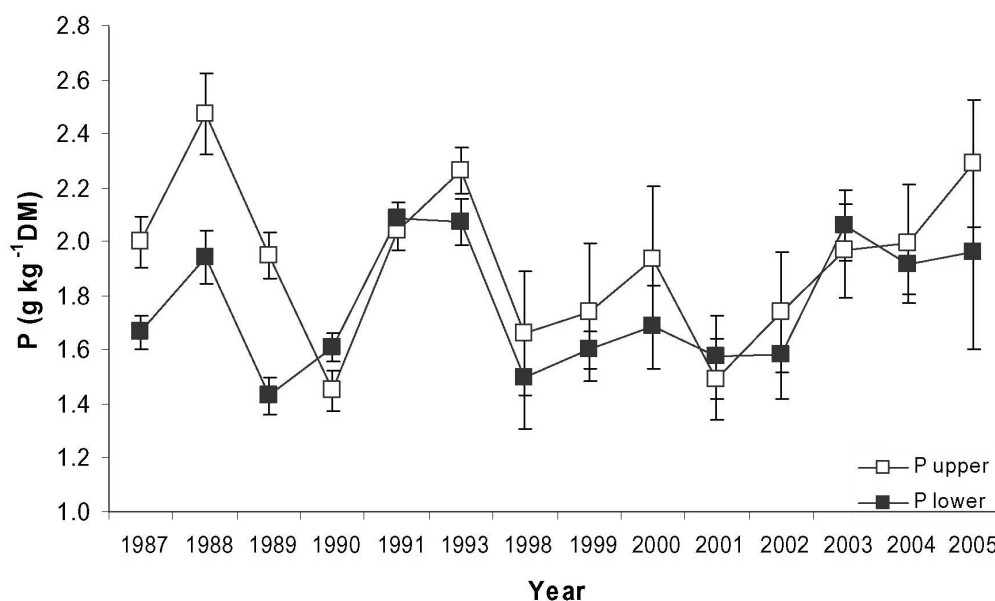


Figure 1. Phosphorus concentration (g kg<sup>-1</sup> dry matter) in 'dehesa' pastures, in the upper and the lower slope zones from 1987 to 2005. Values are means  $\pm$  SE.

The P content of the upper zones was significantly correlated to that observed for the lower zones along the sampling years ( $r = 0.683$ ;  $P = 0.007$ ;  $n = 14$ ).

The P concentration was not significantly ( $P > 0.05$ ) correlated with any climatic variable along the sampling years. When all data were considered, P concentration was significantly correlated with the proportion in dry weight of botanical components in the herbage (Table 1). The P concentration in the herbage was significantly and negatively correlated with the proportion of grasses in both slope zones, and positively with the proportion of legumes in the lower zones and with the proportion of forbs in the upper zones.

Table 1. Pearson correlation coefficients between P content in the upper and lower zones and the proportion and DM production of botanical components (grasses, legumes and forbs) and summary DM production including all species (pasture). Significance level is in brackets.

	Proportion over dry weight			Dry matter production			
	Grasses	Legumes	Forbs	Grasses	Legumes	Forbs	Pasture
P upper	-0.313 (0.000)	0.105 (0.139)	0.236 (0.001)	-0.228 (0.001)	0.146 (0.038)	0.169 (0.016)	0.039 (0.579)
P lower	-0.199 (0.002)	0.209 (0.001)	0.082 (0.208)	-0.204 (0.001)	0.101 (0.119)	0.002 (0.972)	-0.078 (0.227)

The P content in pasture was significantly correlated to available P in soil in 1990 ( $r = 0.838$ ,  $P < 0.000$ ) but not in 1987 or 2005. These results suggest that the P content in pasture is not directly related to the available P in soil. Soils of the study area are P-deficient. This could indicate that differences in soil available P contents among locations or sampling years are not strong enough to provoke differences in P content of herbage. On the other hand, in the three years, the P content in the pasture was negatively significantly ( $P > 0.05$ ) correlated with carbon and organic matter contents in soil and positively with the coarse sand fraction.

In all sampling years, the mean P concentration of pasture was below the phosphorus requirement for adequate cattle nutrition (NRC, 2000). An experiment of phosphoric fertilization conducted on other 'dehesas' has shown that the application of phosphoric rock can give acceptable improvements in biomass production (Ferrera *et al.*, 2006).

## Acknowledgements

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# The influence of organic and mineral fertilizers on fodder quality in NE Romania

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## Abstract

The pastoral area of Romania (4.9 million ha), of which 3.4 million ha are in grassland and 1.5 million ha are hayfields, is an important source of fodder for livestock. This paper reports on the influence of a sustainable management, applied on permanent grasslands at 770 m a.s.l. in Pojorata-Suceava County, and at 820 m a.s.l. in Cosna-Suceava County, on the quality of the fodder obtained. In the experiment at Pojorata, investigations were made on an *Agrostis capillaris* + *Festuca rubra* grassland of the influence of organic fertilizers, at rates of 10-30 Mg ha<sup>-1</sup>, on the background of 30-50 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>. In the experiment at Cosna, the influence of organic fertilization was investigated on a *Nardus stricta* grassland using rates of 20-50 Mg ha<sup>-1</sup> applied each year or every two years, on the content of crude protein, crude fibres, phosphorus and raw ash. The applied fertilization systems resulted in increased fodder yield and CP content, as compared with the unfertilized control, by 14-29% on *Nardus stricta* grassland and by 9-22% on *Agrostis capillaris* + *Festuca rubra* grassland. An improvement in the content of phosphorus in fodder was also found.

Keywords: fodder quality, permanent grasslands, crude protein, crude fibres

## Introduction

The quality of fodder, in terms of its chemical composition, is very important for obtaining high zootechnical productions. The chemical indicators under study here are crude protein, crude fibre, ash, phosphorus and non-nitrogenous extractive substances. The modifications found within these structures are determined by botanical composition, plant anatomo-morphological specific features, vegetation phase at the harvesting period and fertilization level (Vintu *et al.* 1993, Schils *et al.*, 1999; Todorova and Kirilov, 2002; Vuckovic *et al.*, 2005; Poldisek *et al.*, 2007) It is interesting for us to find out if the nature of fertilizers used for fodder production on permanent grasslands, whether as organic or mineral fertilizers, or the frequency of application, may result in significant changes to the chemical composition of fodder, directly influencing its quality.

## Materials and methods

The trials were set up in the mountainous region, where fodder from grasslands is mainly used for dairy cow feeding, on *Nardus stricta* and *Agrostis capillaris* + *Festuca rubra* grasslands. Organic fertilization was used as an experimental factor, with rates varying between 20 and 50 Mg ha<sup>-1</sup>, applied each year or every two years, respectively. Organic fertilizers at rates of 10-30 Mg ha<sup>-1</sup>, were applied each year or every two-three years, in association with mineral fertilizers, at rates of 30-50 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>. Nitrogen was applied totally in spring (30 kg total-N ha<sup>-1</sup>) or split, in spring, and after the first harvesting (30 + 20 kg total-N ha<sup>-1</sup>). The unfermented manure, having a content of 0.42% total-N, 0.19% P<sub>2</sub>O<sub>5</sub>, 0.27% K<sub>2</sub>O and 29% DM, was applied manually very early in spring, at the beginning of the grass growth. Experiments were set up according to the method of randomized blocks with four replicates, as compared with the unfertilized control. We used high manure rates for achieving a rapid diminution of the percentage of *Nardus stricta* species in the vegetal canopy.

For determining the crude protein, we have used the Kjeldahl method, for crude fibre, the Weende method, for total phosphorus, the photometrical method, and ashing for the raw ash. The fodder chemical analyses were carried out on samples taken at the second harvesting cycle. The analysed data represented the average of years 2006-2007.

## Results and discussion

The organic fertilization of *Nardus stricta* grassland, at moderate rates of 20-30 Mg ha<sup>-1</sup> manure, is a means of increasing the content in crude protein (CP) up to 24.7 g kg<sup>-1</sup> DM, as compared with the unfertilized control. The rates of 40-50 Mg ha<sup>-1</sup> have shown the same tendency of CP increase, without great differences as compared with lower rates, but they greatly diminished the percentage of dominant species and led to a DM yield increase by 71-118%, compared with the control. The content of raw ash (RA) has progressively increased once with the increase in the rate of applied organic fertilizer, varying between 71.0 and 86.9 g kg<sup>-1</sup> DM, in the case of annual organic fertilization, compared with only 60.4 g kg<sup>-1</sup> DM in the control. In the case of applying fertilizers once in two years, the changes in the raw ash content were reversed, diminishing as the manure rate increased. The content of crude fibre (CF) was inversely proportional to CP, being highest in the control (280.6 g kg<sup>-1</sup> DM) and it was lowest in the case of fertilization with 40 Mg ha<sup>-1</sup> applied once in two years (203.5 g kg<sup>-1</sup> DM). Phosphorus, an important element for animal feeding, has increased from 1.3 g kg<sup>-1</sup> DM in the control, to 2.4 g kg<sup>-1</sup> DM in the case of fertilization with 50 Mg ha<sup>-1</sup> manure applied every two years (Table 1).

Table 1. Influence of organic fertilization on fodder yield (Mg ha<sup>-1</sup> DM) and chemical composition of *Nardus stricta* grassland (g kg<sup>-1</sup> DM).

Treatments	Mg ha <sup>-1</sup>	CP	RA	CF	Fats	P <sub>total</sub>	OM	ESN
Unfertilized control	2.01	85.0	60.4	280.6	18.5	1.3	939.6	555.5
20 Mg ha <sup>-1</sup> manure applied every year	2.62*	109.7***	71.0	264.3	22.1	1.8*	929.0	533.0
30 Mg ha <sup>-1</sup> manure applied every year	3.04**	106.2***	75.8	275.9	17.7	2.1*	924.2	524.4
40 Mg ha <sup>-1</sup> manure applied every year	3.40***	101.5***	82.4	250.2	22.0	2.3**	917.6	543.9
50 Mg ha <sup>-1</sup> manure applied every year	4.38***	104.7***	86.9	254.4	20.4	2.0*	913.1	533.6
20 Mg ha <sup>-1</sup> manure applied every 2 years	2.46 <sup>ns</sup>	99.8***	80.0	240.4	21.3	1.8*	920.0	558.4
30 Mg ha <sup>-1</sup> manure applied every 2 years	2.89*	107.2***	76.5	236.9	22.4	2.0*	923.5	537.0
40 Mg ha <sup>-1</sup> manure applied every 2 years	2.73*	96.6***	71.4	203.5	17.4	2.0*	928.6	621.1
50 Mg ha <sup>-1</sup> manure applied every 2 years	3.24**	106.4***	75.8	226.4	17.3	2.4**	924.2	574.1

\* =  $P \leq 0.05$ ; \*\* =  $P \leq 0.01$ ; \*\*\* =  $P \leq 0.001$ ; ns = not significant

CP = crude protein, RA = raw ash, CF = crude fibre, P<sub>total</sub> = total phosphorus, OM = organic matter, ESN = extractive substances without nitrogen.

On *Agrostis capillaris*-*Festuca rubra* grasslands, the organic and mineral fertilization has shown a DM increase of 31-48%, as compared with the control, and an increase of CP content from 123.2 g kg<sup>-1</sup> DM in the control, to 150.9 g kg<sup>-1</sup> DM, in the case of fertilization with 20 t ha<sup>-1</sup> manure, applied every two years and 30 + 20 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>. The split-nitrogen application did not result in positive increases of CP content in the second cycle of production, except the treatment with application of 20 t ha<sup>-1</sup> every two years. The content of raw ash from fodder has shown an increase from 94.4 g kg<sup>-1</sup> DM at the control to 113.4 g kg<sup>-1</sup> DM at the fertilization with 30 Mg ha<sup>-1</sup> manure, applied every three years with 30 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>. The phosphorus content has registered increases from 1.6 g kg<sup>-1</sup> DM for the control, to 1.9 g kg<sup>-1</sup> DM, in the case of fertilization with 30 Mg ha<sup>-1</sup> manure every three years with 30 + 20 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>, without being influenced by the split application of nitrogen. In the fertilized variants, the content in crude

fibre varied between 175.9 and 200.6 g kg<sup>-1</sup> DM (20 t ha<sup>-1</sup> manure, applied every two years with 30 + 20 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>, respectively, 30 t ha<sup>-1</sup> manure, applied every three years, with 30 + 20 kg total-N ha<sup>-1</sup> and 30 kg P ha<sup>-1</sup>), while in the control, it was of 170.5 g kg<sup>-1</sup> DM (Table 2).

Table 2. Influence of organic and mineral fertilization on fodder yield (Mg ha<sup>-1</sup> DM) and chemical composition from *Agrostis capillaris-Festuca rubra* grassland (g kg<sup>-1</sup>DM).

Treatments	Mg ha <sup>-1</sup>	CP	RA	CF	Fats	P <sub>total</sub>	OM	ESN
Unfertilized control	3.45	123.2	94.4	170.5	23.6	1.6	915.6	558.3
10 Mg ha <sup>-1</sup> manure applied every year + N <sub>30</sub> P <sub>30</sub>	4.94**	140.4***	100.2	194.2	24.2	1.7 <sup>ns</sup>	899.8	541.1
10 Mg ha <sup>-1</sup> manure applied every year + N <sub>30+20</sub> P <sub>30</sub>	4.51*	133.9***	100.7	195.0	26.2	1.7 <sup>ns</sup>	899.3	554.2
20 Mg ha <sup>-1</sup> manure applied every 2 years + N <sub>30</sub> P <sub>30</sub>	5.12**	146.7***	106.8	186.4	25.6	1.8 <sup>ns</sup>	893.2	534.4
20 Mg ha <sup>-1</sup> manure applied every 2 years + N <sub>30+20</sub> P <sub>30</sub>	4.83*	150.9***	107.4	175.9	25.9	1.8 <sup>ns</sup>	892.6	539.9
30 Mg ha <sup>-1</sup> manure applied every 3 years + N <sub>30</sub> P <sub>30</sub>	4.64*	145.7***	113.4	188.6	27.1	1.7 <sup>ns</sup>	886.6	525.2
30 Mg ha <sup>-1</sup> manure applied every 3 years + N <sub>30+20</sub> P <sub>30</sub>	4.78*	139.4***	106.5	200.6	27.8	1.9*	893.5	525.5

\* = P≤0.05; \*\* = P≤0.01; \*\*\* = P≤0.001; ns = not significant

CP = crude protein, RA = raw ash, CF = crude fibre, P<sub>total</sub> = total phosphorus, OM = organic matter, ESN = extractive substances without nitrogen.

## Conclusions

The fertilization of *Nardus stricta* grassland with 20-50 Mg ha<sup>-1</sup> organic manure has resulted in important changes of fodder chemical composition, greatly improving its quality by increasing the CP content from 85.0 g kg<sup>-1</sup> DM (control) to 109.7 (20 Mg ha<sup>-1</sup> manure applied each year), total phosphorus from 1.3 to 2.4 g kg<sup>-1</sup> DM, and raw ash from 60.4 to 86.9 g kg<sup>-1</sup> DM, and diminishing the content in crude fibre from 280.6 to 203.5 g kg<sup>-1</sup> DM, thus increasing the fodder digestibility.

The changes in fodder quality on *Nardus stricta* grassland were more obvious at application rates of 20-30 Mg ha<sup>-1</sup> manure every year than at using rates of 40-50 Mg ha<sup>-1</sup> applied every two years.

Although on the *Agrostis capillaris-Festuca rubra* grassland there was a better botanical composition, the tested fertilization variants also resulted in improved fodder quality, by increasing the content of CP from 132.2 to 150.9 g kg<sup>-1</sup> DM, of raw ash from 94.4 to 113.4 g kg<sup>-1</sup> DM, and of phosphorus from 1.6 to 1.8-1.9 g kg<sup>-1</sup> DM.

Applying an additional nitrogen fertilizer dose after the first harvest (20 kg total-N ha<sup>-1</sup>) did not result in essential changes in the fodder chemical composition from the second vegetation cycle.

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## **Session 3B**

### **Forage conservation for feed and biomass production**

# The effect of silage making technology on production and quality of milk

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## Abstract

Two silage making technologies were compared using first and second cut timothy-meadow fescue silages in 2003 and 2004. For conventional silage (CS) grass was mown in windrows, wilted for 2-20 h, harvested with precision-chop harvester, treated with formic acid and stored in bunker silo. For round baled silage (BS) grass was mown into a full width swath, wilted for 24-59 h, windrowed, baled without additive, and wrapped. In both years sixteen cows were used in a repeated Latin square experiment with a 2 × 2 factorial arrangement of treatments to evaluate the effects of silage making technology (CS and BS) and cutting time (first and second cut) on milk yield, milk composition, organoleptic quality and renneting properties. In 2004 when weather conditions during harvesting were unfavourable, the use of BS reduced silage intake and subsequently decreased milk yield as compared to CS. However, no adverse effects on milk quality were observed. In 2003 the harvesting conditions were good, and no significant differences in milk production were observed between the technologies.

Keywords: grass harvesting, silage, baling, additive, wilting, milk production

## Introduction

The development in grass harvesting methods aims for labour efficient silage making systems while the effects on silage quality are often ignored. In baling technology, high dry matter (DM) concentration is required for successful preservation and cost effective production. Low water concentration reduces the risks for clostridial fermentation (McDonald *et al.*, 1991) and thus a major part of high DM silages is made without additive. Silage quality may, however, be compromised by yeasts and moulds. An efficient wilting method, such as wide spreading, is needed to speed up drying and to reduce the risks for nutrient losses during wilting. The method may, however, increase soil contamination and, thus, endanger microbial quality of the silage. The effect of quality in low DM grass silage on intake and milk production has been studied extensively (Huhtanen *et al.*, 2003). Regarding high DM silages, data on silage quality risks and responses in milk production are scarcer. The aim of this study was to investigate the effect of harvesting system on nutritive value and hygienic quality of silages and their effects on milk production and quality.

## Materials and methods

Primary and secondary growth of a mixed timothy-meadow fescue sward was used in 2003 (Exp. 1) and 2004 (Exp. 2). For conventional silage (CS), grass was mown with a mower conditioner in windrows, wilted for 2-20 h, harvested with a precision chopper, treated with a formic acid-based additive (formic acid 760 and ammonium formate 55 g kg<sup>-1</sup>), and stored in a bunker silo. For round baled silage (BS), grass was mown with a mower conditioner into a

full width swath, wilted for 24-59 h, windrowed, cut and baled without additive, and wrapped using white 750 mm stretch film with 10 (Exp. 1) or 6 layers (Exp. 2).

In both years, sixteen cows were used in a repeated 4 x 4 Latin square experiment with a 2 x 2 factorial arrangement of treatments to evaluate the effects of silage making technology (CS and BS) and cutting time (first and second cut). The cows were 63 days (Exp. 1) or 57 days (Exp. 2) in lactation. The study consisted of four 21-day periods with a sampling period of the last seven days. Silages were fed *ad libitum* and the daily concentrate portion was 13 kg (multiparous) or 11 kg (primiparous). The concentrate consisted of barley, oats, rape seed meal, dry molassed sugar beet pulp, and mineral-vitamin supplement.

Representative samples of feeds were collected during the sampling period. Milk samples from four consecutive milkings were analysed for protein, fat, and lactose. Samples for milk urea and analysis of milk organoleptic quality and renneting properties were taken from two consecutive milkings. The data was subjected to analysis of variance using the general linear model procedure (PROC GLM) of SAS (1989). The statistical model included the effects of block, cow within block, period and treatment. The treatment effect was separated for effects of the silage technology and cutting time and their interaction.

## Results and discussion

The grass yield was higher in 2004 than in 2003 (5,100 and 4,200 kg in first and second cut vs. 3,300 and 3,100 kg DM ha<sup>-1</sup>). In addition to higher grass yield, rainy weather explains the rather low DM concentration in BS in 2004 (Table 1). Silage pH was higher in BS than in CS in both years, because no additive was used and high DM relatively to bunker silage reduced lactic acid and VFA concentration in BS. The silage ammonia concentration was low in 2003 but lower DM concentration induced higher ammonia production in 2004. The visible deterioration in BS was limited in both years. Instead, problems were observed in aerobic deterioration of CS probably due to slow feeding rate of the silage. This was evidenced by higher mould counts in 2003 and higher clostridia counts in the second cut CS in 2004 compared to BS.

Table 1. Wilting time, dry matter concentration, and quality of the silages (n = 4 for each silage).

Cutting time Silage	Experiment 1 (2003)				Experiment 2 (2004)			
	First cut		Second cut		First cut		Second cut	
	Silo	Bale	Silo	Bale	Silo	Bale	Silo	Bale
Wilting time, h	14	59	2	24	12	25	20	42
Dry matter, g kg <sup>-1</sup>	301	525	349	695	258	386	279	479
pH	4.16	6.01	4.65	6.18	4.17	5.82	4.29	5.92
WSC, g kg <sup>-1</sup> DM	171	175	122	120	120	122	65	71
Lactic acid, g kg <sup>-1</sup> DM	22.6	1.4	19.7	0.9	37.6	14.9	43.9	19.7
Acetic acid, g kg <sup>-1</sup> DM	15.1	8.0	14.0	5.4	16.5	8.2	13.1	8.1
Butyric acid, g kg <sup>-1</sup> DM	0.19	0.23	0.20	0.15	0.60	0.30	1.25	0.70
Ammonia N, g kg <sup>-1</sup> N	28.5	28.7	44.3	13.4	43.5	64.0	70.6	77.4
D-value, g kg <sup>-1</sup> DM	740	736	666	702	715	712	646	634
Yeast, log cfu g <sup>-1</sup>	1.15	2.17	1.87	1.74	1.00	3.57	3.14	2.73
Mould, log cfu g <sup>-1</sup>	2.43	1.39	6.39	2.30	2.41	1.91	2.70	2.45
Clostridial spores, log MPN g <sup>-1</sup>	0.48	0.48	1.17	0.73	0.78	0.88	3.04	1.31

Silo = CS, conventional silage, Bale = BS, bale silage, WSC = water soluble carbohydrates, D-value = digestible organic matter in dry matter, cfu = colony forming unit, MPN = most probable number.

In Exp. 1 in 2003, no significant differences in milk yield were observed between the silage making methods (Table 2) although the use of baling technique for first cut silage decreased

intake compared to CS (interaction  $P < 0.001$ ). In Exp. 2 in 2004, the use of BS reduced both silage intake and milk yield ( $P < 0.001$ ) compared to CS. The decreased intake could not be explained by silage fermentation quality, digestibility, or DM concentration. The effects on intake may be related to differences in silage particle length (in CS 80%  $< 40$  mm, in BS 85%  $> 80$  mm).

Milk protein concentration in Exp. 1 was higher ( $P < 0.01$ ) and the renneting properties were better ( $P < 0.05$ ) in both experiments in cows fed BS than in those fed CS. Milk taste or cheese parameters (Mäki *et al.*, unpublished) were not affected by dietary treatments, suggesting that the microbial quality of silages was not reflected in milk quality. Good milking hygiene probably prevented, e.g., the transfer of clostridia spores to milk.

Table 2. The effect of experimental silages on feed intake, milk production, and milk quality.

Cutting time Silage	First cut		Second cut		SEM	Statistical significance		
	Silo	Bale	Silo	Bale		Silo vs. Bale	Cutting time	Inter- action
<i>Experiment 1 (2003)</i>								
Silage intake, kg DM d <sup>-1</sup>	13.8	13.0	13.0	13.1	0.11	**	**	***
Total intake, kg DM d <sup>-1</sup>	24.7	24.0	24.0	24.1	0.12	*	*	**
Milk, kg d <sup>-1</sup>	38.2	38.0	37.3	37.9	0.21		*	
Fat, g kg <sup>-1</sup>	43.5	42.3	42.0	40.9	0.59		*	
Protein, g kg <sup>-1</sup>	33.1	33.4	32.6	33.1	0.12	**	**	
Curd firmness, A30, mm	26.2	27.1	25.5	27.1	0.61	*		
Milk taste (1-5)	4.21	4.15	4.09	4.16	0.053			
<i>Experiment 2 (2004)</i>								
Silage intake, kg DM d <sup>-1</sup>	14.0	12.9	11.4	9.7	0.17	***	***	
Total intake, kg DM d <sup>-1</sup>	24.9	23.9	22.4	20.8	0.17	***	***	
Milk, kg d <sup>-1</sup>	36.2	34.4	33.4	32.5	0.33	***	***	
Fat, g kg <sup>-1</sup>	45.4	45.2	43.4	42.8	0.62		**	
Protein, g kg <sup>-1</sup>	34.3	34.0	33.1	32.8	0.15		***	
Curd firmness, A30, mm	24.3	28.2	21.8	25.1	1.54	*		
Milk taste (1-5)	4.11	4.10	4.19	4.22	0.035		*	

Statistical significances: \*  $P < 0.05$ ; \*\*  $P < 0.01$ ; \*\*\*  $P < 0.001$ , SEM = standard error of the mean.

## Conclusions

Harvesting conditions affected the nutritive value of high DM, untreated bale silage more than that of additive-treated minimum wilted bunker silage. Harvesting method appeared to affect silage intake irrespective of fermentation quality and digestibility. When the harvesting method requires high DM concentration, the control of silage quality is challenging in unfavourable conditions. In the present experiments, the quality of high DM bale silage did not affect milk quality adversely.

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# Quality and economics of pre-wilted silage made by wide-spreading or by swathing

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## Abstract

When ensiling, it is recommended to wilt the green crop and let it dry to 400-500 g kg<sup>-1</sup> before ensiling it in round bales. During cutting, the mower commonly places the crop in swaths in the field. If the mower places the crop wide-spread in the field instead, it dries faster and in a more controlled way. In this experiment, the wilting process was monitored and silage quality compared when the crop was wilted using the wide-spreading technique versus the swathing technique. When the round baling began 25 h after mowing, the DM in the wide-spread crop was about 400 g kg<sup>-1</sup>, while it was about 280 g kg<sup>-1</sup> in the swathed crop. Butyric acid content was 0.4 g kg<sup>-1</sup> DM in the wide-spread crop and DM losses were 19 g kg<sup>-1</sup>, while these parameters in the swathed crop were 7.3 and 45 g kg<sup>-1</sup>, respectively, showing a better silage quality with the wide-spreading technique. Production costs were also recorded. The extra labour and machinery costs required in the wide-spreading treatment were compensated by faster mowing work and higher density in the bales, resulting in 10% lower total costs for the wide-spreading treatment.

Keywords: silage, quality, pre-wilting

## Introduction

Wilting forage to dry matter (DM) contents as high as 400 g kg<sup>-1</sup> prior to ensiling provides several advantages. It reduces DM losses in bunker, tower and bale ensiling systems. It also increases the forage density and thereby increases harvesting efficiency due to time savings when baling or in filling loader wagons. Another valuable benefit is the elimination of undesirable clostridia growth during fermentation.

The most common way to wilt forage is to let it dry in the swath formed by the mower conditioner (Lingvall, 1995). This method, however, may result in non-uniform forage DM content in the swath (wet in the bottom and dry on top). Insufficient homogenization of such forage can create wet spots in silages, which encourages the growth of spoilage bacteria and can affect the hygienic quality of an entire silo. Therefore, it is of interest to test and compare forage wilted in swaths with forage that is wide-spread and then windrowed prior to ensiling. On the one hand, wide-spreading promotes a more even DM distribution in the crop and shortens wilting time. On the other hand, the wide-spreading method requires an extra pass for windrowing before baling which results in an additional cost and a risk of contaminating the crop with soil during the windrowing operation.

The present study compares the windrowing system with the wide-spreading system mainly in terms of drying efficiency.

## Materials and methods

A 20 ha ley consisting of *Festuca pratensis*, *Phleum pratense*, *Trifolium pratense* and *Taraxacum vulgare* in fresh weight proportions of 50/40/5/5 was cut in the primary growth with a mower conditioner. On one part of the ley the mower produced a wide-spread crop, while it was made into swaths on the other part. The wide spreading was performed with a

front-mounted Pöttinger Novacat 306 FED and a side-mounted Pöttinger Novacat 305 HED on the same tractor, producing a layer of about 10-20 cm thickness. The swathing was performed with a front-mounted Pöttinger Novacat 306 FED and a towed Pöttinger CatNova 3200T with side elevator creating one 'double' swath (1.8 m wide x 0.3 m high). The wide-spread crop was put into windrows after wilting, just prior to round baling with a Pöttinger Eurotop 771 A. Duplicate samples to determine DM contents were taken throughout the wilting period 13 times at the top and bottom layers of the crop separately. Fresh samples were weighed, chopped and mixed and a sub-sample dried in a ventilated drying chamber at 60 °C for 18 h. When one of the two wilting treatments reached the DM content 400 g kg<sup>-1</sup>, estimated by a quick micro-wave analysis, the round baling started for both treatments using a Welger 220 Profi Combibaler that presses and wraps in the same machine. In total 60 bales were made in three blocks. Bale sampling was performed with a 4-cm diameter stainless steel corer. Six horizontal cores of 40 cm depth were extracted from three randomly selected bales per block of ten bales, producing 18 samples (nine bales from swathed and nine bales from wide-spread crop sections). All silage samples were analysed for DM, water soluble carbohydrates (WSC), pH, ammonium-N, fatty acids and ethanol using standard methods. Analysis of variance in a completely randomized block design was used to evaluate the effects of wilting technique on silage quality (SAS, 2002).

## Results

The crop was cut in the afternoon. Shortly thereafter, it received 2-4 mm rain. As a result of the rain, the DM content dropped, slightly less in the swathed crop compared with the wide-spread since it was not exposed to the rain in the same way. The weather during the following day was a mixture of sun and cloud without rain. At 1600 h the wide-spread treatment reached 400 g DM kg<sup>-1</sup> and the harvest started. The DM development in the two treatments is presented in Figure 1.

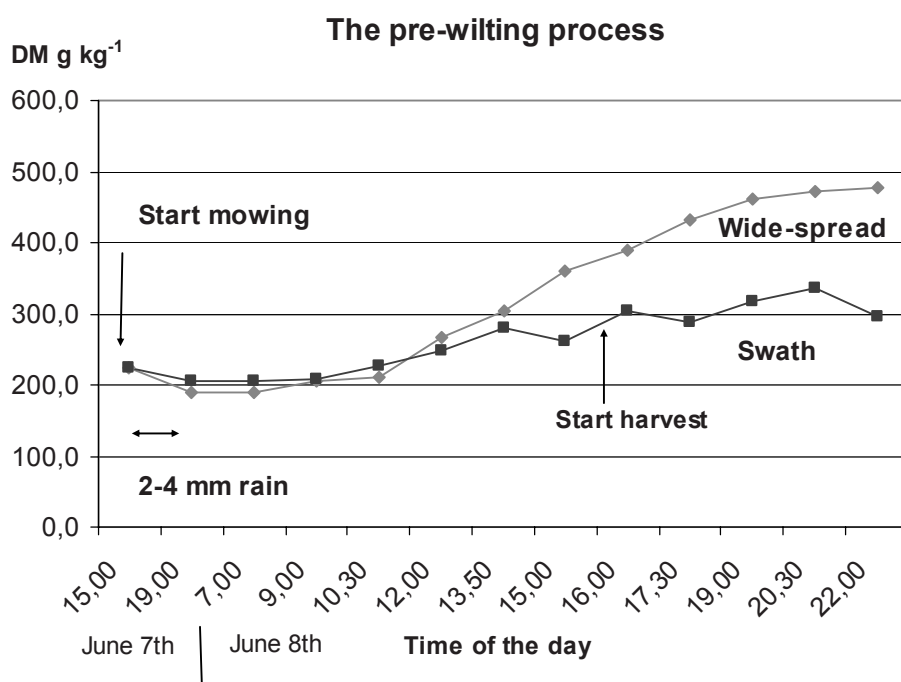


Figure 1. Degree of dryness (g kg<sup>-1</sup> DM) during the wilting process after mowing at 1,500 hours until 2,200 hours the following day. The round baling started at 1,600 hours the second day.



At the start of harvest, the swathed crop had reached about 280 g DM kg<sup>-1</sup> with a variation from about 400 g kg<sup>-1</sup> on the top to about 200 g kg<sup>-1</sup> in the bottom with a coefficient of variation (CV) of DM analysis during the second day of 181. The DM variation in the wide-spread treatment was significantly lower with a CV of the samples during the same period of 21.

The resulting silages differed significantly in quality (Table 1). The wide-spreading gave a good silage quality, while the swathing produced silage with twice as much ammonia and an unwanted high concentration of butyric acid. The DM loss was twice as high in the swathed silage. The bale density (kg DM per bale) was 30% higher in the wide-spread treatment.

Table 1. Silage quality, density and DM losses in round bales produced from green crop, wilted wide-spread or in swaths for 25 hours. SD = standard deviation.

	Wide-spread		Swathed		Significance <i>P</i> <
	mean (N = 30)	SD	mean (N = 30)	SD	
DM, g kg <sup>-1</sup>	416	24.2	285	17.2	0.001
pH	5.2	0.09	4.8	0.13	0.001
NH <sub>3</sub> -N, g kg <sup>-1</sup> total N	65.1	11.38	109.6	14.87	0.001
<i>In dry matter:</i>					
WSC, g kg <sup>-1</sup>	86.0	11.92	43.2	15.33	0.001
Lactic acid, g kg <sup>-1</sup>	15.4	4.69	39.6	10.74	0.001
Acetic acid, g kg <sup>-1</sup>	3.6	0.90	8.8	2.89	0.001
Butyric acid, g kg <sup>-1</sup>	0.4	0.08	7.3	4.35	0.001
Ethanol, g kg <sup>-1</sup>	12.2	3.35	14.7	2.54	0.001
2,3-butandiol, g kg <sup>-1</sup>	3.1	0.56	13.9	3.21	0.001
Density, kg DM m <sup>-3</sup>	181	12.8	137	9.7	
DM loss, g kg <sup>-1</sup>	1.9	0.61	4.5	1.04	

The economical aspects of the wilting systems were studied by recording time spent and fuel consumption during all phases of the trial. The wide-spreading system had 10% lower costs despite the extra windrowing work. This was due in part to faster mowing (4.49 ha hr<sup>-1</sup> vs. 3.35 ha h<sup>-1</sup>), but mainly to more effective baling of the crop (170 vs. 117 kg DM min<sup>-1</sup>) and fewer bales because of the higher DM content (341 vs. 269 kg DM bale<sup>-1</sup>).

## Discussion and conclusions

The better silage quality achieved with wide spreading probably was due to the higher DM content. Silage quality with pre-wilting in swaths to the same DM was not studied. However, by wide-spreading the crop in a layer of approximately 10-20 cm thickness instead of a 30 cm swath, the wilting process takes place faster, even when the crop is exposed to light rainfall during the wilting process. This can be decisive for how early it is possible to start the making of silage, thereby decreasing the risk of exposure to further rainfall. In the present experiment the swathed crop would have to stay another night and probably the next half-day to reach the same DM content as the wide-spread crop. The use of wide-spreading technique is often criticised because it requires more labour, including an extra tractor pass across the field to windrow the crop prior to the silage making. However, the economical effectiveness of the wide-spreading technique can be superior when it results in higher DM content.

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# Loader wagon compared to metered chopper for forage harvest

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## Abstract

A comparison was made between five loader wagons (LW) of size 31-42 m<sup>3</sup> and a metered chopper (MC) in terms of their relative functional performance. In the second cut in 2005, mown grass was windrowed from a width of 8.5 m just before harvesting. The dry matter content was 173-194 g kg<sup>-1</sup>. The mean chop length for the LWs was 90 mm and for the MC 31 mm. The loader wagons had different knife distances, between 34 and 45 mm, but no significant difference in chop length. The mean fuel consumption was 0.24 l Mg<sup>-1</sup> for the LWs and 0.31 l Mg<sup>-1</sup> for the MC. The value for the MC does not include fuel needed for towing the wagon, because the wagon was towed with another tractor at the side of the MC. The energy consumption for the cutting was 0.57 kWh Mg<sup>-1</sup> for the LWs and 0.94 kWh Mg<sup>-1</sup> in the MC. Pilot trials with LW indicated that increased driving speed can reduce chop length, and that drier grass can reduce fuel consumption.

Keywords: grass harvesting, loader wagon, metered chop harvester, energy consumption, chop length

## Introduction

Technical solutions are significant inputs which influence the cost and value of silage and which significantly impact on choices made by farmers (Forristal and O'Kiely 2005). Self-loading forage wagons have increased in size and capacity, and have become fast and efficient harvesting machines. The total capacity of the loader-wagon method depends much on the distance from field to silo, and to some extent on how the wagons fill their loading space. With more general attention being paid to energy matters, the fuel consumption of the harvesting method is of interest. Another property of interest is the chop length, which can affect silage quality and the performance of feeding equipment. The objective of this study was to measure these properties in different loader wagons and to compare them with a metered chopper.

## Material and methods

Five brands of loader wagons (LW) with size 31-42 m<sup>3</sup> were compared in harvesting trials and other measurements in 2005. The project also included studies of practical usability, which are not reported here. The harvesting trials were done on an even 20 ha field in a second cut of a sward of mixed timothy (*Phleum pratense*) and meadow fescue (*Festuca pratensis*). There was a trailed JF 1350 metered-chop harvester (MC) as a reference machine. The field trials were done in a randomized block design. Three replicates, i.e. loads, were made with each harvesting machine. The grass was mown with a 3.2 m mower conditioner. Just before harvesting, it was windrowed from a width of 8.5 m with a centre-delivery windrower. Power take-off torque (HBM torque sensor), PTO speed (HBM), fuel consumption (Pierburg PLU 116 H sensor), and driving distance were measured in the harvesting trials. The sensors and data collection were coupled in a Valtra 8950 tractor. All loads were weighed. Three dry matter and three chop length samples, minimum 500 g each, were taken from each load. The chop lengths of the herbage were determined by hand-separating a 100 g sample into 8 categories (0-20, 21-40, 41-60, 61-80, 81-100, 101-160, 161-220, and > 220 mm). The herbage in each length category was dried, weighed, and the

percentage distribution in each of the categories calculated. The load densities of the wagons was calculated as a measure of how well the wagons filled up and utilized their loading space, by dividing the load weights with the wagon volume. The results of chop length, filling degree, and fuel consumption were tested with analysis of variance. Due to a sensor breakage, there is only one measured load regarding PTO power and fuel consumption for MC, and therefore the MC could not be included in the analyses of variance of PTO power and fuel consumption.

## Results and discussion

The harvest conditions were rainy and the forage was wet, and the dry matter (DM) content ranged from 173 to 194 g kg<sup>-1</sup>. The differences in chop length between the LWs tended to follow the differences in their knife distances. However, the differences in chop length among the five LWs were not statistically significant (Table 1). However, in all LWs the chop lengths differed significantly from the MC chop length ( $P < 0.01$ ).

Table 1. Chop length and knife distance of loader wagons and metered-chop harvester (MC).

Machine and model	Knife distance (mm)	Mean length (mm)	SD	In relation to MC
Pöttinger Torro 5700	35	82 a	11.04	2.65
Bergmann Shuttle 880K	34	83 a	22.48	2.68
Strautmann Giga-Vitesse I	40	94 a	19.40	3.03
Krone 5 XL	40	95 a	31.25	3.06
Claas Quantum 5500P-18	45	98 a	25.79	3.16
LW mean		90 a		2.92
JF 1350 (MC)		31 b	4.10	1.00

Values with different letters are significantly different ( $P < 0.01$ ).

An additional pilot experiment indicated that increased driving speed of LWs, i.e. increased mass flow of forage through the knives, can reduce chop length. Increased speed from 5 to 12 km h<sup>-1</sup> with the Krone LW shortened the mean chop length from 79 to 44 mm. The Pöttinger LW showed the same trend, although not as systematically as in Krone (Table 2). Because this was a pilot trial with only one replicate, the essential conclusion is the trends, not the differences among LW makes or absolute numbers. The mass flow through the knives also can be increased by windrowing swaths together.

Table 2. Driving speed and chop length of LW in pilot trial. Krone's knife distance was 40 mm, Pöttinger's 35 mm.

Krone 6XL		Pöttinger Torro 5700	
Driving speed (km h <sup>-1</sup> )	Mean length (mm)	Driving speed (km h <sup>-1</sup> )	Mean length (mm)
5	79	3	96
8	68	7	117
12	44	11	85

The highest load densities were reached with the MC and the Strautmann, Pöttinger, and Krone LWs; there were no significant differences among these LWs (Table 3). The Claas had a significantly lower load density compared with the MC, but not compared with the other LWs. The wet grass was probably disadvantageous for the largest LW, Bergmann. Its load density was lowest, 72% of Strautmann's. The reason could be that the tractor was too small, or the narrow swath width, dimensioned to fit the narrower pickups of the other wagons. In a later pilot trial (one replicate) with drier grass, DM 280 g kg<sup>-1</sup>, Bergmann filled itself as well as the other LWs.

Table 3. Load density and fuel consumption with loader wagons and metered-chop harvester (MC).

Machine and model	Load density		Relative	Fuel consumption	
	kg m <sup>-3</sup>	SD		l t <sup>-1</sup> grass fresh weight	SD
Bergmann Shuttle 880K	269 a	40.3	72	0.26 ab	0.041
Claas Quantum 5500P-18	319 ab	33.5	86	0.21 a	0.014
Krone 5 XL	350 bc	11.8	94	0.24 ab	0.053
Pöttinger Torro 5700	360 bc	28.7	97	0.21 a	0.020
Strautmann Giga-Vitesse I	373 bc	54.8	100	0.27 b	0.021
JF 1350 (MC)	399 c	49.0	107	0.31*	

Values with different letters are significantly different ( $P < 0.05$ ).

\*not directly comparable to the loader wagons; the tractor pulled only the chopper, no wagon. MC n = 1.

The PTO power requirements in Table 4 apply only to the driving speeds used, and the speeds were different for MC and LW. To get a measure of the energy requirement which is comparable among the machines, the PTO power requirements have to be put in relation to the mass flow through the machine and the driving time. The resulting parameter is PTO energy per harvested Mg (tonne) of grass. This was 0.57 kWh Mg<sup>-1</sup> for the LWs and 0.94 kWh Mg<sup>-1</sup> for the MC. This means that MC is a 68% greater energy user than LW in absolute numbers. However, when taking into account that the MC with its average chop length of 31 mm does about three times more cutting work than the LW with its average chop length of 90 mm, the MC is more energy efficient in relation to the amount of cutting work. Short chop is desirable when the herbage is much wilted, to provide better chances for good consolidation and fermentation. Some feeding equipment also require short chop. Unfortunately the PTO power and energy results for the MC are from only one load, due to a sensor breakage. The PTO results for LW are from one load each of Bergmann, Claas, Krone and Pöttinger.

Table 4. Power requirement and capacity for loader wagon (LW) and metered-chop harvester (MC).

	LW	MC
Average speed in windrow, km h <sup>-1</sup>	8.1	5.0
Average PTO power, kW	84.6	113
Max. PTO power, kW	129	145
Energy requirement for the cutting, kWh Mg <sup>-1</sup>	0.57	0.94
Forage, DM g kg <sup>-1</sup>	182	177
Forage/load, kg	11963	8540
Average loading rate, Mg min <sup>-1</sup>	2.4	2.0
Load density, kg m <sup>-3</sup>	334	399

## Conclusions

There were no significant differences in chop length among the five LWs. The LWs made significantly – three times – longer chop than the MC. The MC used 68% more energy for the cutting than the LWs in absolute numbers. On the other hand, the MC was more energy efficient than the LWs in relation to the amount of cutting work. According to pilot trials, the chop length of LWs tended to decrease with increased mass flow through the knives. With wet forage there were differences in how the machines filled the loading space. Pilot trials with drier forage indicated smaller differences in this respect.

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# Quality and aerobic stability of big-bale silage treated with bacterial inoculants containing *Lactobacillus buchneri*

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## Abstract

An experiment was carried out to examine silage fermentation, aerobic stability and nutritive value in pre-wilted grass silage, which was ensiled in big bales either without additive (control) or with two bacterial silage additives containing homo- and heterofermentative lactic acid bacteria (treatments K1 and K2). Silages were examined for dry matter, pH, chemical and microbiological composition and aerobic stability. The addition of inoculants increased the lactic acid and decreased the butyric acid concentration ( $P < 0.05$ ) in silage. Both additive treatments resulted also in improvement ( $P < 0.05$ ) of aerobic stability of silage that was accompanied by the reduction of yeast and mould counts in silages. The silages were offered *ad libitum* with concentrate supplementation to twenty-seven heifers blocked into three weight groups for a period of 91-92 days. Silage dry matter intakes and live-weight gains among heifers fed different silages were not different.

Keywords: aerobic stability, bacterial inoculant, grass silage, *Lactobacillus buchneri*

## Introduction

It is well known that addition of biological additives enhances the fermentation process during ensiling and, thereby, increases the possibilities to obtain a good fermentation quality and a high digestibility of nutritive components in grass silage. One of the consequences of increasing the proportion of lactic acid is the negative impact on aerobic stability. It is accepted that well preserved, high quality silages, inoculated only with homofermentative lactic acid bacteria (LAB) are more prone to aerobic spoilage than untreated (Weinberg *et al.*, 1993). Aerobic deterioration of silage can affect both the efficiency of nutrient utilization and its hygienic quality. The improvement of aerobic stability is important in case of ensiling plant material in big bales with high dry matter content. The recent developments have focused on the use of heterofermentative LAB, particularly *Lactobacillus buchneri* (Driehuis *et al.*, 2001), which produces acetic acid inhibiting the yeast development, that may improve silage stability after bales are opened (Danner *et al.*, 2003). The aim of this study was to investigate the influence of two bacterial additives containing the homo- and heterofermentative LAB on the fermentation quality, aerobic stability and nutritive value of grass silage in big bales.

## Materials and methods

Silages were made from a meadow sward composed of 80% grasses (*Poa pratensis*, *Alopecurus pratensis*, *Dactylis glomerata*, *Arrhenatherum elatius*, *Lolium perenne*) and 20% weeds and herbs. The meadow was cut three times a year in 2005 and 2006; first at full heading of *Dactylis glomerata* and then at nine-week intervals. The herbage was mown with a rotary mower-conditioner and pre-wilted to a dry matter (DM) concentration of approximately 400-450 g kg<sup>-1</sup>. The meadow herbage was ensiled either without treatment (control silage), with the addition of commercial inoculant containing: *Enterococcus faecium* M74, *Lactobacillus casei*, *Lactobacillus plantarum*, *Pediococcus* spp., *Lactobacillus buchneri*

(Polmass SA; Poland; treatment K1) or with another commercial inoculant containing: *Lactobacillus plantarum* K, *Lactobacillus plantarum* C, *Lactobacillus brevis*, *Lactobacillus buchneri* (IBPRS, Poland; treatment K2). Both additives were applied at the rate of 2 l t<sup>-1</sup> fresh herbage by spraying during bale-rolling in a flex bale chamber baler. The big bales (about 400 kg/bale) were wrapped in four layers of stretch film after transport to their place of storage. In January of each next year the bales were opened for feeding and sampling. Two feeding trials, one in 2005 and one in 2006 were performed. Twenty-seven heifers (mean initial live weight of 215 ± 22 kg in 2005 and 198 ± 35 kg in 2006) were blocked into three groups of nine according to live weight. The heifers (black-white breed) were offered silage, either with additives (treatment K1 or treatment K2) or without the additives *ad libitum* and supplemented with a controlled amount of concentrate for a period of 91 (2005) and 92 (2006) days. Treated and untreated silages from first, second and third cut were fed during three 30 day-long consecutive periods. The daily feed intake and live weight gains for every trial were determined. During the feeding experiment representative samples of fresh silage (3 samples per treatment from every cut) were taken for determination of: DM (oven method), pH, ammonia-N concentration (distillation method), organic acids (the enzymatic method), nutritive components (NIRS technique), mould and yeast counts. Aerobic stability of silages was analysed by monitoring the temperature changes in samples (3 replications) placed in boxes in aerobic conditions (ambient temperature about 21 °C) for 12 days. Stability was measured as the time necessary to increase silage temperature by 1 °C over air ambient temperature. Data concerning the chemical composition of silage were analysed using analysis of variance in ANOVA 3. Silage intakes and animal performance were compared using ANOVA 2. Differences between treatments were tested using the Student's t-test.

## Results and discussion

Mean DM contents in examined silages were typical for pre-wilted grass silage. The concentration of organic acids in silage DM depended mainly on the treatment. Inoculant-treated silages displayed higher ( $P < 0.05$ ) lactic acid and lower butyric acid concentrations than the untreated silage. Concentrations of acetic acid and ammonia-N were lower in K1-treated silage compared with the control treatment ( $P < 0.05$ ). In general, the addition of bacterial inoculants containing homo- and hetero-fermentative LAB improved fermentation quality. The silages prepared with inoculants obtained higher ( $P < 0.05$ ) Flieg-Zimmer scores than control silage (Table 1). The addition of additives increased ( $P < 0.05$ ) aerobic stability of silage from 6.5 days (control) to 9.4 days (K1 treatment). The aerobic spoilage of silage depended also on the time of harvest (cut) and the year. The silages from the third cut and the second year were more prone to aerobic spoilage than the remaining feeds. The inoculant-treated silages had lower ( $P < 0.05$ ) yeast and mould counts than the untreated silage. The reason for this effect is an anaerobic degradation of lactic acid to acetic acid (Oude Elferink *et al.*, 2001) that inhibits yeast and mould development (Danner *et al.*, 2003). Inoculant treatment had an evident impact on the nutritive value of the feeds. The total protein, WSC and NEL content were higher ( $P < 0.05$ ) and crude fibre was lower ( $P < 0.05$ ) in inoculated silages than in control silage. The differences ( $P < 0.05$ ) were found in nutritive component concentrations among silages produced from swards harvested at different occasions (cuts). When fed to heifers, DM intake of silage and animal performance among silage treatments were not different. In spite of high concentration of acetic acid in inoculant-treated silages, even in the case of K1 silage, a decline of silage intake was not observed. However, no differences between live weights among animals were found. Heifers offered the tested silage in 2006 showed higher daily live weight gain than those offered the silage in 2005 ( $P < 0.05$ ).



Table 1. Chemical composition, aerobic stability and nutritive value of silages.

Examined parameters	Treatment (T)			Cut (C)			SEM	Year (Y)		SEM	Significance <sup>2)</sup>			
	Control	K1	K2	I	II	III		2005	2006		T	C	Y	TxC
DM (g kg <sup>-1</sup> )	472	422	477	463 <sup>a</sup>	513 <sup>a</sup>	395 <sup>b</sup>	5.094	467	447	4.159	ns	**	ns	ns
pH	5.01 <sup>a</sup>	4.49 <sup>b</sup>	4.74 <sup>ab</sup>	4.56 <sup>b</sup>	4.86 <sup>a</sup>	4.83 <sup>ab</sup>	0.082	4.91 <sup>a</sup>	4.59 <sup>b</sup>	0.067	**	*	**	ns
NH <sub>3</sub> -N (g kg <sup>-1</sup> total N)	48.6 <sup>a</sup>	31.8 <sup>b</sup>	44.7 <sup>ab</sup>	41.9 <sup>a</sup>	27.9 <sup>b</sup>	55.3 <sup>a</sup>	4.722	40.5	42.9	3.856	*	**	ns	ns
Lactic acid (g kg <sup>-1</sup> DM)	23.0 <sup>b</sup>	39.0 <sup>a</sup>	37.1 <sup>a</sup>	36.4 <sup>a</sup>	25.1 <sup>b</sup>	37.7 <sup>a</sup>	2.088	34.7	31.4	1.705	**	**	ns	ns
Acetic acid (g kg <sup>-1</sup> DM)	10.2 <sup>b</sup>	14.5 <sup>a</sup>	10.1 <sup>b</sup>	12.8	10.9	10.9	0.872	7.4 <sup>b</sup>	15.6 <sup>a</sup>	0.712	**	ns	**	*
Butyric acid (g kg <sup>-1</sup> DM)	1.2 <sup>a</sup>	0.4 <sup>b</sup>	0.3 <sup>b</sup>	0.4	0.8	0.7	0.115	0.5 <sup>b</sup>	0.8 <sup>a</sup>	0.094	**	ns	*	ns
Flieg-Zimmer score <sup>1)</sup>	55.8 <sup>b</sup>	86.3 <sup>a</sup>	89.9 <sup>a</sup>	83.4 <sup>a</sup>	67.0 <sup>b</sup>	81.6 <sup>a</sup>	3.211	86.7 <sup>a</sup>	68.0 <sup>b</sup>	2.622	**	**	**	ns
Yeast (log <sub>10</sub> cfu g <sup>-1</sup> FM)	2.53 <sup>a</sup>	1.54 <sup>b</sup>	1.59 <sup>b</sup>	1.79 <sup>b</sup>	1.93 <sup>ab</sup>	1.94 <sup>a</sup>	0.041	1.81 <sup>b</sup>	1.97 <sup>a</sup>	0.033	**	*	**	ns
Moulds (log <sub>10</sub> cfu g <sup>-1</sup> FM)	2.99 <sup>a</sup>	1.81 <sup>b</sup>	1.89 <sup>b</sup>	1.96 <sup>b</sup>	2.34 <sup>a</sup>	2.40 <sup>a</sup>	0.053	1.94 <sup>b</sup>	2.52 <sup>a</sup>	0.043	**	**	**	**
Stability (days)	6.50 <sup>c</sup>	9.39 <sup>a</sup>	7.61 <sup>b</sup>	8.44 <sup>a</sup>	8.17 <sup>a</sup>	6.89 <sup>b</sup>	0.350	8.63 <sup>a</sup>	7.04 <sup>b</sup>	0.286	**	**	**	*
Total protein (g kg <sup>-1</sup> DM)	127 <sup>b</sup>	143 <sup>a</sup>	137 <sup>a</sup>	126 <sup>b</sup>	129 <sup>b</sup>	152 <sup>a</sup>	2.618	133	138	2.138	**	**	ns	**
Crude fibre (g kg <sup>-1</sup> DM)	264 <sup>a</sup>	257 <sup>ab</sup>	255 <sup>b</sup>	256 <sup>b</sup>	278 <sup>a</sup>	242 <sup>c</sup>	2.323	254 <sup>b</sup>	263 <sup>a</sup>	1.897	*	**	**	ns
WSC (g kg <sup>-1</sup> DM)	118 <sup>b</sup>	125 <sup>a</sup>	125 <sup>ab</sup>	133 <sup>a</sup>	112 <sup>c</sup>	123 <sup>b</sup>	2.314	132 <sup>a</sup>	113 <sup>b</sup>	1.889	*	**	**	ns
NEL (MJ kg <sup>-1</sup> DM)	5.16 <sup>b</sup>	5.19 <sup>ab</sup>	5.25 <sup>a</sup>	5.26 <sup>a</sup>	5.10 <sup>b</sup>	5.24 <sup>a</sup>	0.025	5.19	5.21	0.020	*	**	ns	ns
DM Intake (kg d <sup>-1</sup> )	6.27	5.88	6.32	-	-	-	0.216	5.94	6.37	0.176	ns	-	ns	-
Daily gains (kg)	0.80	0.87	0.82	-	-	-	0.033	0.76 <sup>b</sup>	0.90 <sup>a</sup>	0.027	ns	-	**	-

<sup>1)</sup> 100-score scale where 81-100 scores – very good, 61-80- good, 41-60 – satisfactory, 20-40 – poor, 0-20 – bad quality.

<sup>2)</sup> There was a TxY and TxCxY interaction ( $P < 0.05$ ) only for yeast count and stability;

SEM – standard error of means; Values in rows with different superscripts differ significantly ( $P < 0.05$ ); ns – not significant;

\* and \*\* – significance of main effects and interactions at  $P < 0.05$  and  $P < 0.01$ , respectively.

## Conclusions

It is concluded that the addition of bacterial inoculants containing homo- and hetero-fermentative LAB improved the fermentation quality and aerobic stability of grass silage made in big bales. The additive treatments did not increase silage intake and had no effect on daily live weight gain of heifers.

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# Effect of conservation method on fatty acid composition in model grass silage

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## Abstract

An experiment was conducted to investigate the effects of wilting, additive and their interactions on the fatty acid (FA) composition of grass silage. The crop was timothy (*Phleum pratense* L., cv. Grindstad), and the additives were Proens<sup>TM</sup> (60-66% formic acid, 25-30% propionic acid), the bacterial inoculant Siloferm® Plus (*Pediococcus acidilactici*, *Lactobacillus plantarum*) and water (control). The wilted material reached a dry matter content of 336 g kg<sup>-1</sup> at the first cut, and 350 g kg<sup>-1</sup> at the second cut. The silos used were 1,700 ml glass jars with screw tops, provided with water seals. Neither wilting nor additives had any major effect on the FA proportions. The only significant ( $P < 0.05$ ) differences found were in the concentrations of C16:0 and C18:3. Silage treated with bacterial inoculant contained higher proportions of C16:0 than silage treated with acid (161 vs. 153 g kg<sup>-1</sup> FA), and a significantly lower concentration of C18:3 than silage treated with either acid or water (591 vs. 615 and 604 g kg<sup>-1</sup> FA). These results indicate that we would be spared the conflict between the losses of FAs during wilting and the positive effects which comes out of wilting, such as reduced losses of nutrients through effluents and a higher feed intake by the animals.

Keywords: silage, acid additive, bacterial inoculant, wilting, fatty acid composition

## Introduction

Oxidation during field wilting has been identified as one of the main reasons for losses of polyunsaturated fatty acids (PUFA) in herbage (Dewhurst *et al.*, 2003). Use of additives may also affect these variables, but reported findings in this respect are conflicting. For instance, Dewhurst and King (1998) found that although additives had major effects on overall fermentation parameters, their effects on the levels and proportions of FAs in grass silage were relatively minor. In contrast, Warren *et al.* (2002) found that use of formic acid or bacterial inoculants decreased total FA concentrations, and Boufaïed *et al.* (2003) found that use of a bacterial inoculant resulted in significant declines in the concentrations of C18:3 and total FAs in grass silage and haylage. The objective of this study was to investigate the main and interactive effects of wilting and additive on the FA composition of grass silage made from timothy.

## Materials and methods

The experiment was conducted in 2004 at the research centre Rönneby, Swedish University of Agricultural Sciences, Umeå, Sweden (63°45'N; 20°17'E). The crop used was timothy (*Phleum pratense* L., cv. Grindstad), and the additives were Proens<sup>TM</sup> (60-66% formic acid, 25-30% propionic acid, Perstorp Inc., Perstorp, Sweden), and the bacterial inoculant Siloferm® Plus (*Pediococcus acidilactici*, *Lactobacillus plantarum*; Medipharm Inc., Kågeröd, Sweden). Both the first (14 June) and the second (29 July) cuts were included in the study and the grass was mainly in the sheath-elongation developmental stage when harvested. The fertilization level was 80 kg N ha<sup>-1</sup> for the first cut and 40 kg N ha<sup>-1</sup> for the



second cut. Immediately after cutting, the fresh material was either ensiled within 2 h of cutting or placed on a tarpaulin for wilting. The wilted material reached a dry matter content of 336 g kg<sup>-1</sup> at the first cut, and 350 g kg<sup>-1</sup> at the second cut. Due to logistical problems, the bacterial inoculant was not added to the unwilted material in the first cut. All additives were diluted with water (1:1) and thereafter the mixture was added at a rate of 8 ml kg<sup>-1</sup>. The silos used were 1,700 ml glass jars with screw tops, provided with water seals to release gas. The silos were stored in a dark room at 20 °C for 92 days. Once the silos had been emptied, the silages were stored at -20 °C for 5-6 months until analysis. Lipids were extracted directly from the frozen samples using a slightly modified version of the method described by Folch *et al.* (1957), and then methylated according to the method described by Appelqvist (1968). There were two replicates of each treatment. The GLM procedure in SAS was used for statistical evaluation, and harvest time, additive and wilt were used as fixed effects in the statistical model.

## Results and discussion

All silages were well preserved except for the wilted control silage from the first cut, which had considerably higher levels of butyric acid and ammonium, and a somewhat higher pH than the other silages. All FAs investigated in this study were significantly affected by harvest time, but not by wilting, and there were no major overall differences between the effects of different additives on FA composition (Table 1). Statistically significant ( $P < 0.05$ ), but not substantial, between-additive differences were found in the proportions of only two FAs. Silage treated with the bacterial inoculant had a higher proportion of C16:0 ( $P < 0.05$ ) than silage treated with acid, and use of the bacterial inoculant also resulted in significantly lower proportions of C18:3.

Table 1. Fatty acid (g kg<sup>-1</sup> fatty acids) and crude fat (g kg<sup>-1</sup> DM) concentrations in the different silages.

Harvest time	Treatment	u/w <sup>1</sup>	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	Other <sup>2</sup>	Crude fat	DM (g kg <sup>-1</sup> )
14/6	Acid	U	147.8	16.8	11.5	15.4	156.4	639.0	13.3	3.83	235.7
	Inoculant	U	---	---	---	---	---	---	---	---	---
	Water	U	153.0	12.9	11.6	24.2	163.0	626.3	11.7	3.62	238.8
	Acid	W	149.0	17.1	11.8	15.5	156.4	637.5	12.8	3.73	304.3
	Inoculant	W	159.1	15.1	12.0	16.4	173.0	610.7	13.8	3.68	296.4
	Water	W	151.3	15.1	11.5	18.7	163.0	626.3	14.2	4.06	300.7
	Acid	U	154.0	16.8	12.6	22.0	176.0	601.0	20.4	3.22	169.6
	Inoculant	U	164.2	18.2	13.1	24.2	170.3	578.7	31.6	3.08	170.5
	Water	U	165.5	18.7	17.4	21.8	167.4	581.0	28.4	2.35	165.8
29/7	Acid	W	161.5	17.2	12.4	22.9	186.6	583.0	16.5	2.87	370.4
	Inoculant	W	160.1	17.5	13.1	22.0	179.0	583.2	25.1	2.69	357.4
	Water	W	159.0	16.5	12.6	22.3	187.4	582.5	19.9	2.87	353.4
Source of variation											
Harvest time (H)			**	**	*	**	**	**	**	**	
Additive (A)			**	ns	ns	ns	ns	**	ns	ns	
Wilting (W)			ns	ns	ns	ns	ns	ns	ns	ns	
H*A			ns	*	ns	*	*	ns	ns	**	
H*W			ns	ns	ns	ns	*	ns	ns	ns	
A*W			*	ns	ns	ns	ns	ns	ns	ns	

<sup>1</sup>Unwilted or wilted. <sup>2</sup>Other identified fatty acids: C14:0, C20:0, C20:4 (n-6), C22:0, C22:5 (n-3), C24:0 (not all of them in all samples).

No significant differences in the concentrations of C16:1, C18:0, C18:1 or C18:2 appeared to be associated with either the degree of wilting or use of the different additives. Neither were there any significant differences in crude fat concentration between silages subjected to the different degrees of wilting, or between silages treated with different additives (Table 1).

Wilting did not affect the proportions of FAs as expected. Previous studies (Dewhurst and King, 1998; Boufaïed *et al.*, 2003; Elgersma *et al.*, 2003) have found significantly lower concentrations of C18:3 in wilted than in unwilted material, but in the present study there were no such differences. However, in these three studies the material was wilted for longer periods or to higher DM contents than the grass used in the present study, where the first cut was wilted overnight at rather cool temperatures (10-12 °C) and the second cut for just 6 h. Thus, the differences in FA concentrations may be due to differences in the duration and other parameters of processes catalysed by plant enzymes after cutting (Elgersma *et al.*, 2003), which were allowed to continue for a longer period in two of the previous studies. Moreover, Chow *et al.* (2004) compared three different varieties of perennial ryegrass and found differences in their susceptibility to FA oxidation, indicating that there are differences among varieties. Two of the varieties the cited authors used were affected by ensiling, which resulted in lower concentrations of C18:3. However, the C18:3 concentrations in the third variety were not affected to any major extent by either wilting or ensiling. The timothy used in this study is probably a variety that is affected relatively little by the wilting process. It was, however, affected by the ensiling process, even though the differences between the fresh material and the silages were numerically small (figures not shown).

## Conclusions

When wilted for a short period and to a moderate DM content, the wilting process did not significantly affect the proportions of FAs in *Phleum pratense* L., cv. Grindstad. In addition, the different additives tested in this study had no major effect on the proportions of FAs in silage.

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# Fatty acids in fresh and artificially dried grass

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## Abstract

It is known that during wilting of cut herbage the concentration of fatty acids (FA) changes and that silage contains less unsaturated FA than fresh grass, but information on changes in FA during rapid drying and during storage of dried forage is lacking. This study compared the FA pattern of six samples of fresh grass measured just prior to artificial drying in a commercial drying plant, with that of the dried products after one and seven months of storage. Three grams of cut material of each fresh herbage sample and three grams of dried product were used for lipid extraction and methylation. The fatty acids methyl esters (FAME) were analysed on a Varian GC equipped with a flame ionization detector. The concentration of  $\alpha$ -linolenic acid (C18:3) in fresh grass ranged from 14.3 to 22.9 g kg<sup>-1</sup> DM. In the dried product there was a decline, ranging from circa 25 to 45% of the C18:3 from the DM. FA concentrations did not change in the dried product after storage. The FA profile was similar in fresh and dried material. C18:3 was the main FA, contributing 65.3 to 74.2% of the total amount of FAME. Free FA were found in very low concentrations compared with esterified FA, both in fresh and artificially dried grass. The outcomes of this pilot study indicated that artificially dried grass may be more beneficial than ensiled grass from the viewpoint of maintaining unsaturated FA in winter diets for ruminants.

Keywords: dried grass, dehydration, grass pellets, fatty acid, feed quality

## Introduction

The content of fatty acids (FA) in fresh and conserved forage such as silage has recently come into focus in ruminant nutrition, due to increasing interest in dairy products with a modified FA composition. Milk FA composition is largely determined by FA intake (i.e., total FA content and composition of individual FA) with the feed. The lipid fraction in leaves of herbs and grasses ranges from 30 to 100 g kg<sup>-1</sup> dry matter (DM). Fresh grass contains a high proportion (0.50-0.75) of total FA content as  $\alpha$ -linolenic acid. In detached plants immediately after cutting, metabolism of plant cells continues and there is still activity of the enzymes. Continuation of processes of respiration and proteolysis are best known, but lipolysis also occurs.

For instance, Dewhurst and King (1998) found a reduction of 30% in the content of total FA, and of 40% in the content of C18:3 during wilting prior to ensiling. After hay making, the total FA content declined by over 50%, with a proportionally higher loss of C18:3 (Doreau and Poncet, 2000). A similar decline in FA content was reported in haylage (700 g DM kg<sup>-1</sup>; Elgersma *et al.*, 2003).

Reducing FA losses due to lipolysis and oxidation during wilting prior to conservation are important strategies to manipulate precursors for conjugated linoleic acid (CLA) in milk. Information on changes in FA during rapid drying and subsequent storage of dried forage is lacking. Dehydration of forage has the purpose of maintaining as much as possible of the contents of protein, carotenes, vitamins, calcium, and phosphorus that are present in the fresh product and would largely be lost with natural drying. Forage dehydration leads to a fast decrease in free water content, lowering the moisture content below 8%. The fast drying helps

to prevent both respiration of plant tissues and epiphytic bacteria activity. If in dried grass products the FA would be better preserved than in silage or hay, inclusion in winter diets could increase FA intake. This study compared the FA pattern of six samples of fresh grass measured just prior to artificial drying in a commercial drying installation with that of the dried products after one and seven months of storage.

## Materials and methods

Six grass samples were taken on 14 Nov 2006 from a commercial drying plant prior to dehydration. The grass had been cut on different commercial farms the previous day. These farms were all located on sandy soil, in the North-East region of The Netherlands. The species composition was predominantly perennial ryegrass. Per sample, 3 g of cut material was sub-sampled and extracted immediately. All samples were processed similarly in the factory, i.e., dried at 900 °C during 1-2 minutes. Thereafter, sample 1 was baled and samples 2-6 were ground and pelleted. These dried grass products were then sampled and stored at room temperature in the dark. On 21 December and 19 June, 3 g was used for extraction. Part was used to analyse free FA prior to methylation. Extracts were methylated and FA methyl esters were analysed on a Carlo Erba 5,200 gas chromatograph, equipped with a flame ionization detector as reported earlier (Elgersma *et al.*, 2003).

## Results and discussion

FA concentrations in fresh grass ranged from 18.7 to 30.9 g kg<sup>-1</sup> DM, sample 4 being highest (Figure 1). This grass lot was harvested at a low herbage yield (540 kg DM ha<sup>-1</sup>) and had probably had a shorter regrowth period than the other samples, where yield ranged from 1,450 (sample 2) to 2,590 kg DM ha<sup>-1</sup> (sample 3). Earlier, we (Elgersma *et al.*, 2003) compared the composition of fresh perennial ryegrass with that of pre-wilted ensiled material, without additives, for six cultivars (pH 5.8, > 700 g DM kg<sup>-1</sup>, < 30 g NH<sub>3</sub> kg<sup>-1</sup> DM) harvested after 25 days of regrowth with a yield of approximately 2000 kg DM ha<sup>-1</sup> and measured on average 28.6 g FA kg<sup>-1</sup> DM in fresh grass, which is in line with the current data.

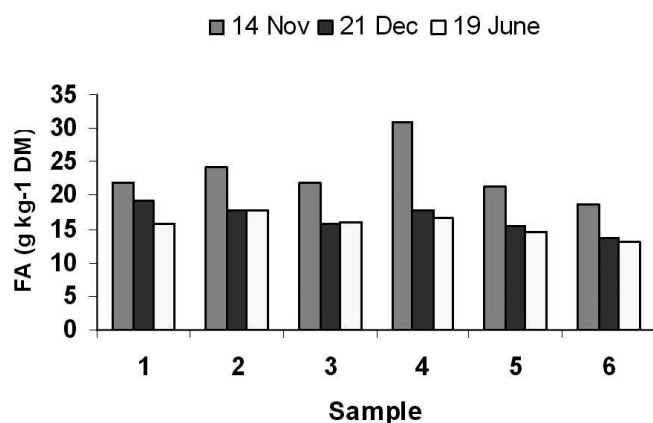


Figure 1. The total concentration of fatty acids (FA) in six samples of fresh cut grass on 14 November 2006, and of dehydrated grass products (bales in sample 1, pellets in samples 2-6) on 21 December 2006 and after six months of storage on 19 June 2007.

After drying, the FA concentrations declined to 13.6 to 19.3 g kg<sup>-1</sup> DM in December, and remained at that level after another 6 months of storage (13.2 to 17.8 g kg<sup>-1</sup> DM in June; Figure 1). In contrast, after ensiling the FA concentration declined from 28.6 in fresh grass to

9.9 g kg<sup>-1</sup> DM in silage (Elgersma *et al.*, 2003), which is a much stronger decline than found in this study after dehydration. There was no evidence from this pilot study for an effect of baling or pelleting on FA.

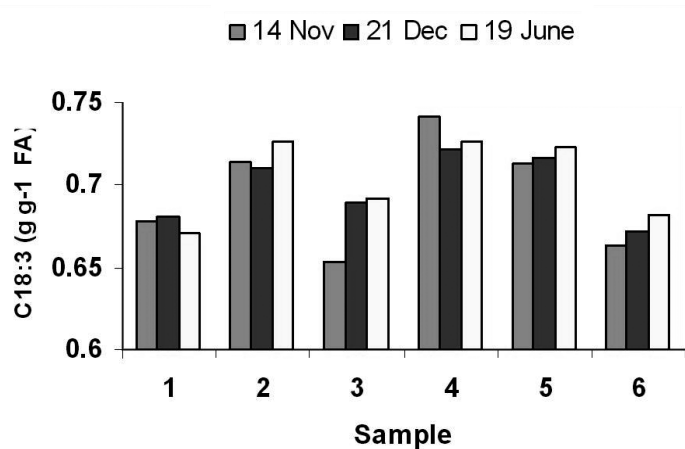


Figure 2. The proportion of  $\alpha$ -linolenic acid, C18:3, in six samples of fresh cut grass on 14 November 2006, and of dehydrated grass products (bales in sample 1, pellets in samples 2-6) on 21 December 2006 and after six months of storage on 19 June 2007.

The proportions of FA were not affected by drying or storage. The proportion of  $\alpha$ -linolenic acid (C18:3) ranged from 0.65 to 0.74 of total FA (Figure 2). Linoleic (C18:2) and palmitic acid (C16:0) had both proportions of 0.14 of total FA (not shown). In our earlier ensiling study (Elgersma *et al.*, 2003), proportions of FA were not affected either (the proportion of C18:3 declined from 0.69 to 0.67).

Free FA contributed less than 0.07 to total FA in both fresh and dried grass, which is in line with earlier findings (Elgersma *et al.*, 2003) on fresh grass. However, after ensiling substantial amounts of free FA were found (0.27-0.73).

## Conclusions

C18:3 was the main FA in fresh grass, with proportions of 0.65 to 0.74 of total FA. The concentration of FA in the fresh grass ranged from 19 to 31 g kg<sup>-1</sup> DM. After dehydration the concentrations of FA decreased with 25 to 45%. The FA profile was similar in fresh and dehydrated grass. FA concentrations in dehydrated grass did not change after storage for up to seven months. More FA were preserved in dehydrated grass compared with hay or silage.

## Acknowledgments

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# Effects of climate and cutting delays on timeliness losses in silage harvest

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## Abstract

This study examined timeliness costs in harvest of forage for milk production. Timeliness costs express the quality and/or quantity losses occurring in the harvested crop when a machine operation is performed at a non-optimal time or with limited capacity. Choice of machinery capacity is an offset between high machine costs due to large machines, and timeliness costs due to low machine capacity. The method used to investigate annual variations in timeliness losses was to construct a model combining DM production of grass-clover forage at different harvesting dates with the economic value of the forage depending on its energy and crude protein content. For forage production in central Sweden (Uppsala area) the timeliness factor was 60 SEK ha<sup>-1</sup> for each day's delay in first harvest from the optimal harvest date. Corresponding values for second and third harvests were 23 and 14 SEK ha<sup>-1</sup> day<sup>-1</sup>, respectively. For southern Sweden, timeliness factors for harvests 1-3 were 80, 28 and 20 SEK ha<sup>-1</sup> day<sup>-1</sup>. The magnitude of timeliness losses varied greatly between years.

Keywords: forage, timeliness, harvest capacity, machinery, milk production

## Introduction

This paper deals with the economic consequences of delays in the timing of forage harvest. Costs incurred from quality and/or quantity losses when harvest is not performed at the optimal time or with limited capacity of the equipment are called timeliness costs.

Seasonal variations in climate are one of the most important constraints on forage productivity. Due to climatic factors, grassland production may vary considerably between years and also within a growing season (Herrmann *et al.*, 2005). An initial study by Gunnarsson *et al.* (2005) showed that forage yield and crop development had great influence on the timeliness costs. Consequently, timeliness losses show great variations between years with variations in climatic conditions.

A calculation model was constructed for estimating timeliness losses for forage harvest. When the time of harvest is delayed from the optimum time, two factors arise that influence the value of the harvest and consequently the timeliness losses: DM yield increases with time and the nutrient content decreases with time. The model therefore consisted of two parts, one of which calculated the DM production of grass-clover forage at different harvest dates using a forage growth model created by Torssell *et al.* (1982) and Torssell and Kornher (1983). The other part of the model calculated the change in forage value caused by changes in energy and crude protein content of the forage.

## Materials and methods

In addition to a number of model parameters specific for the sward and site (Torssell *et al.*, 1982), the model used for the forage yield calculation requires the climate parameters temperature, incoming radiation, precipitation and evaporation. Daily increase in DM yield is

calculated as the product of the current amount of biomass and the relative growth rate  $R_s$ . The initial amount of biomass and the initial  $R_s$  at the start of each growing period account for the influence of botanical composition and management practices such as nitrogen fertilization, number of harvests per season and geographical location. The initial values of  $R_s$  and the initial amount of biomass used in this study were taken from a model of the forage-ruminant system called PCVALL, also a result of the FOPROQ model (Fagerberg *et al.*, 1990).

The value of the forage was calculated using a method developed by Gunnarsson *et al.* (2005):

(a) Regression analyses of field experiments (Fagerberg *et al.*, 1990) were used to calculate harvest days and protein contents corresponding to two specified energy contents of the forage.

(b) The economic value of forages of different nutrient quality was determined by making fodder plans and calculating fodder costs and milk yields. By setting the same value on the total fodder cost for each fodder plan, the value of each forage quality could be calculated.

(c) The difference in economic value between the two forage qualities and the number of days between the two harvest dates were used to calculate the daily change in economic value.

The optimal time to harvest the forage was decided by finding the harvest dates of each harvest that maximize the total value (in € ha<sup>-1</sup>) of all harvests. The objective function describing the total maximum value (denoted  $Z$ ) of the three harvests is described by (formula 1):

$$Z_{tot} = M1 \times V1 + M2 \times V2 + M3 \times V3 \quad (1)$$

where  $M$  denotes the yield in kg DM ha<sup>-1</sup> and  $V$  denotes the value of the forage in € (kg DM)<sup>-1</sup> for first (1), second (2) and third (3) harvest. Delays in first harvest have consequences on the yield of the second and third harvests. Similarly, delays in the second harvest influence the third harvest. By choosing the harvest date for every harvest that resulted in the highest value of the sum of all three harvests, the fact that delays of a harvest also influence the following harvests was taken into account. It was assumed that all harvests were performed on the dates calculated as optimal, e.g. when the harvest dates of second or third harvests were optimal, it was assumed that the preceding harvest was cut on the optimal day.

The timeliness factor, i.e. daily change in value (€ ha<sup>-1</sup> day<sup>-1</sup>), was calculated as the difference between the total value of the harvest at the optimal date and the total value of the harvest seven days after the optimal date.

The calculations were made for three cuts per season at two sites in Sweden (Uppsala in central Sweden and Malmö in the south). The calculations were repeated for a 10-year period using daily climate data from Uppsala for the period 1978-1987 and from Malmö for the period 1984-1993.



## Results and discussion

For southern Sweden, the timeliness factor was 80 SEK ha<sup>-1</sup> day<sup>-1</sup> for the first harvest. Corresponding values for second and third harvest were 28 and 20 SEK ha<sup>-1</sup> day<sup>-1</sup>, respectively. For central Sweden the timeliness factors were calculated to 60, 23, and 14 SEK ha<sup>-1</sup> day<sup>-1</sup> for the first, second and third harvest (Figure 1). Thus timeliness factors were higher in the south of Sweden. This was mainly due to higher yields, as total yields were on average about 25% higher in the south compared with central Sweden.

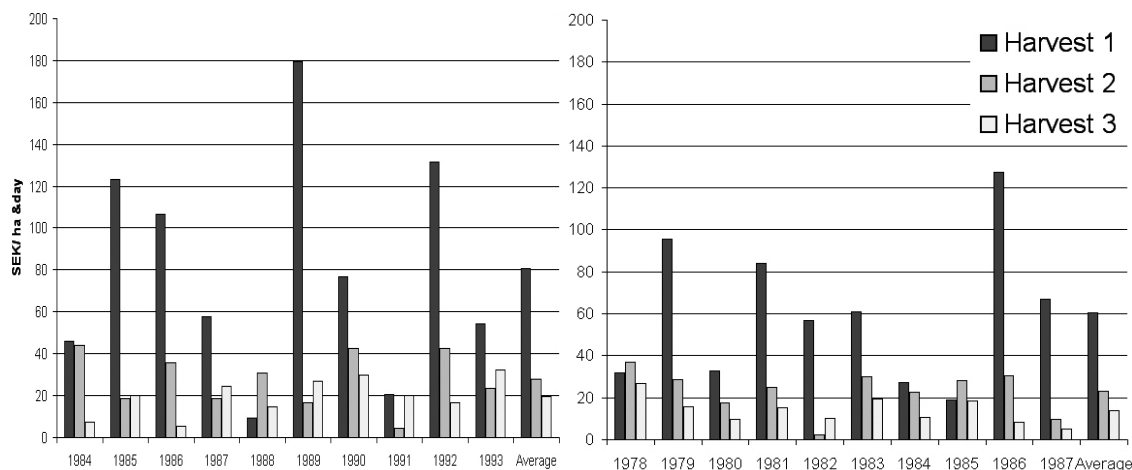


Figure 1. Timeliness factors (SEK ha<sup>-1</sup> day<sup>-1</sup>) for southern Sweden (left) and central Sweden (right).

As a consequence of faster development of the forage crop during the first growth period, timeliness factors were higher for the first harvest compared with the regrowth.

Furthermore, the timeliness factors showed a large variation between years. For example, for the ten years studied, the timeliness factors for southern Sweden varied between 10 and 180 SEK ha<sup>-1</sup> day<sup>-1</sup> for the first harvest.

The total timeliness costs for harvesting a specific area are dependent on when the harvest starts and how long the harvest lasts. The next step will be to use the timeliness factors to quantify the timeliness costs for the forage harvest on farms with varying harvest capacity and harvest system.

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# Effects of a multi-strain inoculant on the fermentation characteristics of lucerne silage

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## Abstract

An effective biological additive contains a range of specifically selected strains of bacteria designed to dominate and direct the fermentation of silage. The multi-strain inoculant (Microsil Extra Plus (MEP), Medipharm, Sweden) was tested for its ability to reduce pH, and improve the fermentation profile and aerobic stability of lucerne (*Medicago sativa*) silage. Lucerne at 340 g kg<sup>-1</sup> dry matter (DM) from five fields was untreated or treated with MEP at 1.5 x 10<sup>5</sup> colony forming units g<sup>-1</sup> fresh forage before ensiling in 0.7 and 3-litre silos. Treatments were replicated three times. Addition of MEP resulted in a more rapid reduction of pH after 3 days (4.79 vs. 5.28;  $P < 0.01$ ) and 97 days of ensiling (4.53 vs. 4.97;  $P < 0.01$ ). Silage prepared with MEP had a higher concentration of lactic acid (26 vs. 20 g kg<sup>-1</sup> DM;  $P < 0.01$ ) and a lower concentration of butyric acid (1.0 vs. 5.1 g kg<sup>-1</sup> DM;  $P < 0.05$ ) and ammonia-N (41 vs. 63 g kg<sup>-1</sup> total N;  $P < 0.01$ ). The MEP also reduced fermentation losses (61 vs. 85 g kg<sup>-1</sup> DM). While MEP silage had a marginally better aerobic stability, both treated and untreated silage were aerobically stable for more than six days.

Keywords: lucerne silage, inoculant, fermentation, aerobic stability

## Introduction

Lucerne (*Medicago sativa*) is commonly used for silage, but it is generally considered difficult to ensile because of a low concentration of water soluble carbohydrates (WSC) and a high buffering capacity (BC) (McDonald *et al.*, 1991). Work on legume silage is timely because of its natural place in organic- and low-input production-systems (Jarvis *et al.*, 1996). Inoculants containing lactic acid bacteria (LAB) are used as silage additives to improve conservation efficiency. The homofermentative (HOM) strains *Lactobacillus plantarum*, *Enterococcus faecium* and *Pediococcus* spp. are used because of their efficient production of lactic acid from WSC in the crop and rapid decrease in pH (Weinberg and Muck, 1996). Other LAB, such as the HET *Lactobacillus buchneri*, produce high concentrations of acetic acid, which reduces fungal growth and thus preserves silage susceptible to spoilage upon exposure to air (Driehuis *et al.*, 1999; Weinberg *et al.*, 2002; Ohlsson *et al.*, 2006). However, most HET LAB are not as competitive as HOM LAB in the silage environment that contains a large number of other micro-organisms (Weinberg and Muck, 1996), justifying the combined use of HOM and HET LAB in a silage inoculant.

The objective was to study the effect of the multi-strained Microsil Extra Plus (MEP) containing HOM and HET LAB on the reduction of pH, fermentation pattern and aerobic stability in lucerne silage.

## Materials and methods

Four cultivars of lucerne from five fields, i.e., 'Europe', 'Zydrune', 'Verko' and 2 x 'Birute', were cut at the initial flowering stage and subsequently wilted to 340 g kg<sup>-1</sup> and chopped at 2 cm. Herbage was either left untreated or treated with MEP before ensiling in 0.7 and 3-litre silos. The MEP contained *Enterococcus faecium*, *Lactobacillus casei*, *Lactobacillus planta-*

rum, *Lactobacillus buchneri* and *Pediococcus pentosaceus*. The MEP was dissolved in 4 litres of water  $t^{-1}$  fresh forage at  $1.5 \times 10^5$  colony-forming units  $g^{-1}$  fresh forage. The same volume of distilled water was used for the untreated control. Herbage in each silo was packed manually to approximately  $170 \text{ kg DM m}^{-3}$ . Herbage composition was determined from at least three forage samples from each field directly after spraying and at the time of filling the silos. The 0.7-litre silos were opened after three days of ensiling to determine pH. The 3-litre silos were opened after 97 days of ensiling to determine silage chemical composition, fermentation parameters and aerobic stability. The five fields and two silage additive treatments were used as factors and replicated three times in a randomized complete block design. The GLM procedure was used in the SAS Statistical package, version 8.2 (SAS Institute Inc., Cary, NC, USA).

## Results and discussion

Lucerne silage often contains higher than desired concentrations of butyric acid, which should be less than  $3 \text{ g kg}^{-1}$  DM in high quality silage. The combination of high BC, and low concentrations of DM, WSC and nitrate of lucerne herbage for silage in this study (Table 1) deemed it more sensitive to extensive proteolysis and high concentrations of butyric acid (Tabacco *et al.*, 2004).

Table 1. Chemical composition and buffering capacity of lucerne herbage.

Parameters	n <sup>1</sup>	Herbage	SEM <sup>2</sup>
Dry matter, $g \text{ kg}^{-1}$	30	340	2.36
Crude protein, $g \text{ kg}^{-1}$ DM	30	203	2.29
Water soluble carbohydrates, $g \text{ kg}^{-1}$ DM	30	49	2.31
Buffering capacity, mE $\text{kg}^{-1}$ DM	15	570	20.3
Nitrate, $g \text{ kg}^{-1}$ DM	15	1.3	0.16

<sup>1</sup>Number of observations; <sup>2</sup>Standard error of a mean.

One benefit of MEP-treated silage was related to a more rapid acidification. The inoculant accelerated the reduction in pH after three days of ensiling ( $P < 0.01$ ). Also, MEP silage had a significantly lower pH ( $P < 0.01$ ) after 97 days of ensilage (Table 2).

Table 2. Chemical composition and fermentation parameters of lucerne silage. Each treatment mean was based on 15 observations.

Measured parameters	Control	MEP treated	SEM <sup>1</sup>	Sign. <sup>2</sup>
Dry matter, DM, $g \text{ kg}^{-1}$	326	332	0.79	ns
Crude protein, $g \text{ kg}^{-1}$ DM	207	214	1.82	*
Water soluble carbohydrates, $g \text{ kg}^{-1}$ DM	10	10	0.43	ns
Neutral detergent fibre, $g \text{ kg}^{-1}$ DM	448	426	2.34	**
Lactic acid, $g \text{ kg}^{-1}$ DM	20	26	0.90	**
Acetic acid, $g \text{ kg}^{-1}$ DM	24	23	1.10	ns
Butyric acid, $g \text{ kg}^{-1}$ DM	5.1	1.0	1.26	*
Ammonia-N, $g \text{ kg}^{-1}$ total N	63	41	0.70	**
pH after 97 days	4.97	4.53	0.0028	**
pH after 3 days	5.28	4.79	0.0018	**
Losses, $g \text{ kg}^{-1}$ DM	85	61	4.46	**

<sup>1</sup>Standard error of a treatment mean; <sup>2</sup>Statistical significance where ns = not significant, \* =  $P < 0.05$  and \*\* =  $P < 0.01$  respectively.

The MEP-treated silage produced more lactic acid but less butyric acid and ammonia-N than the control silage. Thus, the fermentation process was more homolactic in the treated compared with untreated silage. However, proteolysis and fermentation losses were more extensive in the control silage.

The aerobic stability was good for both treatments, > 144 h, but marginally better for MEP treated silage (Figure 1). The acetic acid concentration did not differ between treatments, possibly explaining the similar results in aerobic stability (Table 2 and Figure 1).

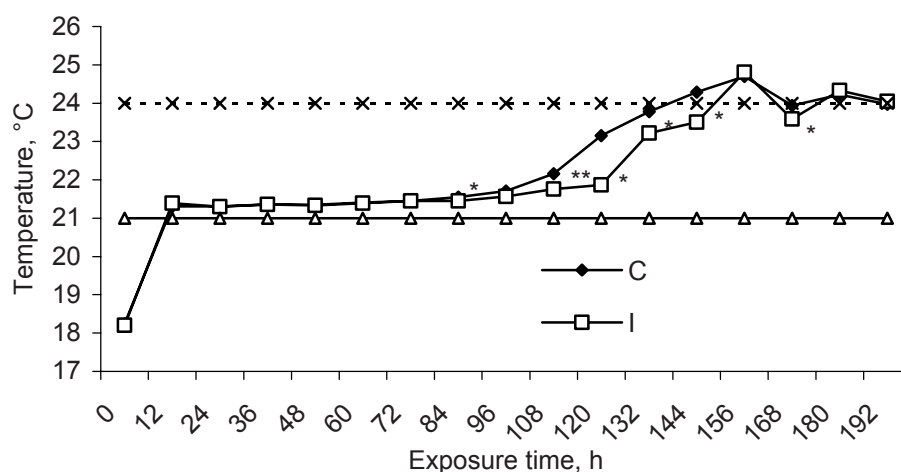


Figure1. Aerobic stability of MEP (I) treated and untreated (C) lucerne silage.

## Conclusions

The MEP-inoculated silage positively influenced the fermentation, resulting in increased concentration of lactic acid and decreased concentrations of butyric acid and ammonia-N and reduced DM losses. Both treatments resulted in silage that was aerobically stable for more than 144 h.

## Acknowledgements

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# Precision chopping or rotor cutting and its influence on ensiling capacity and silage fermentation

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## Abstract

Two self-loading wagon systems were studied during two years. A Pöttinger Jumbo 7200 L rotor cutter wagon (P) of 72 m<sup>3</sup> was compared with Taarup/Sahlström precision chopping wagons (TS) of 37 m<sup>3</sup> in 2004 and 49 m<sup>3</sup> in 2005. The intention was to evaluate harvesting and ensiling efficiencies as well as fermentation characteristics of the forage. Results revealed no differences in silage quality between loading systems as well as no effect on final silage density. The pH, lactic acid and ammonia-N were related to silage DM but all silages were of high quality. Differences between systems in load weight and loading capacity were related to wagon volume, crop laceration and compaction technique. Tonnes of forage DM loaded per hour and fuel consumption showed a strong dependence of crop DM and transport distance. Loading capacities 325 and 467 kg DM min<sup>-1</sup> in P were significantly higher in both years than in TS 40//50 (206 and 322 kg DM min<sup>-1</sup>). Similarly, ensiling capacity was higher in P (5.2/12.4 ton DM h<sup>-1</sup>) compared with TS40/50 (3.6/9.2 ton DM h<sup>-1</sup>). It can be concluded that these results favour the rotor cutting systems, as feeding trials in a later part of the study did not detect any differences in animal performance when silages were fed to dairy cows.

Keywords: dry matter, fermentation, self-loading wagon, silage quality, loading capacity

## Introduction

Mechanical processing of harvested forage is an important procedure in silage-making. Nowadays, there are two main harvesting systems where the crop is precision chopped or processed by a rotor through cutting knives. As both systems have a different technical solutions for processing forages it is of interest to compare these systems in terms of their impact on silage quality and harvesting efficiency. An earlier study showed that precision chopping lacerates the crop to a greater extent than rotor cutting. This increases fermentation rates as defined by a more rapid decrease in pH and an increased production of lactic and acetic acids (Pauly and Lingvall, 1999). Lindell *et al.* (1970) found negative effects of 20 and 80 mm chop length on fermentation characteristics and losses compared with 5 mm length. In addition, precision chopping gives a presumption of better forage homogenization than rotor cutting due to a shorter particle length of the forage. This is important as precision chopping can improve distribution of silage additive in the crop and reduce the problems with wet spots in forage mass that can encourage the growth of spoilage microflora.

## Materials and methods

A clover-grass sward (2004, second cut) and a pure grass sward (2005, first cut) were cut by a Taarup 4032 C mower/conditioner and wilted in a 120-130 cm wide swath for a target dry matter content of 400 g kg<sup>-1</sup>. The forage crops were collected either by a Pöttinger Jumbo 7200 L rotor cutter wagon (P) of 72 m<sup>3</sup> load volume with a combined cutter and loading technique, or Taarup/Sahlström precision chopping wagons of 37 m<sup>3</sup> load volume in 2004 (TS 40) and 49 m<sup>3</sup> in 2005 (TS 50). During loading, the swathes were randomly distributed between loading systems to diminish variations in botanical composition and DM content.



Every wagon was weighed and ten samples per load were taken for DM determination. Loading and transport times and speed were measured for each load separately. The transport distance was estimated. Fuel consumption was measured by filling the tractors at the start of the experiment and then refilling them at the end. The 2004 crop (526 g NDF and 10.3 MJ kg<sup>-1</sup> DM) was preserved in pressed bag silos, and the 2005 crop (494 g NDF and 11.5 MJ kg<sup>-1</sup> DM) in two 6 x 20 m steel silos using similar loading/unloading equipment. Silage density was measured and silage samples were taken during feeding for analysis of silage quality parameters. Five samples per system were taken to determine the distribution of particle length. The ensiling period lasted for five to seven months. Statistical evaluation of loading systems was done separately for each year.

## Results and discussion

Unfavourable weather made it impossible to wilt the crop to 400 g DM in harvest 2004. The crop DM difference between years influenced the fermentation characteristics of silages (Table 1). A lower crop DM in 2004 resulted in intensive fermentation producing more lactic acid (98 vs. 30 g kg<sup>-1</sup> DM), reducing silage pH more than in the silage from 2005. Nevertheless, silages from both years were well fermented resulting in no butyric acid production and acceptable ammonia-N formation. Differences in fermentation pattern were not detected between the loading systems, despite the longer particle length in the P system (Table 2). Arvidsson and Lingvall (2005) identified this tendency in a similar field and laboratory experiment in bunker silos. Table 1 also demonstrates the effect of crop DM on the loading and ensiling efficiency of both loading wagons. A higher crop DM in 2005 made it easier to densely load the crop into the wagon and, thereby, loading weight was increased resulting in improved loading and ensiling capacities of both wagons. The higher load weight of TS50 in 2005 was also related to a larger load volume than TS40 in 2004.

Table 1. Technical results comparing rotor cutting (Pöttinger rotor cutter wagon) and precision chopping (Taarup/Sahlström precision chopping wagons). Values for 2004 and 2005 within a row with different superscripts differ at  $P < 0.05$ .

Harvest*	2004		2005	
Crop yield, kg DM ha <sup>-1</sup>	6.900		4.600	
Wagon	Pöttinger	Taarup TS 40	Pöttinger	Taarup TS 50
<i>Silage composition</i>				
DM, g kg <sup>-1</sup>	241 <sup>a</sup>	225 <sup>a</sup>	399 <sup>a</sup>	404 <sup>a</sup>
pH	4.0 <sup>a</sup>	4.0 <sup>a</sup>	4.4 <sup>a</sup>	4.4 <sup>a</sup>
Butyric acid, g kg <sup>-1</sup> DM	< 0.7	< 0.7	< 0.2	< 0.2
NH <sub>3</sub> -N, g kg <sup>-1</sup> tot-N	83 <sup>a</sup>	78 <sup>a</sup>	48 <sup>a</sup>	45 <sup>b</sup>
Transport distance, km	5.4	5.4	1.8	1.8
Loading volume, m <sup>3</sup>	72	37	72	49
Loading time, min load <sup>-1</sup>	11 <sup>a</sup>	12 <sup>a</sup>	12 <sup>a</sup>	14 <sup>b</sup>
Transport + unloading, min	30 <sup>a</sup>	29 <sup>a</sup>	15 <sup>a</sup>	14 <sup>a</sup>
Load weight, kg DM	3572 <sup>a</sup>	2477 <sup>b</sup>	5598 <sup>a</sup>	4309 <sup>b</sup>
<i>Capacity</i>				
Loaded, kg DM min <sup>-1</sup>	325 <sup>a</sup>	206 <sup>b</sup>	467 <sup>a</sup>	322 <sup>b</sup>
Ensiled, DM hour <sup>-1</sup>	5.2 <sup>a</sup>	3.6 <sup>b</sup>	12.4 <sup>a</sup>	9.2 <sup>b</sup>
Fuel used, L ton <sup>-1</sup> DM	6.0	7.2	2.4	2.5
<i>Relative number</i>				
Density after ensiling kg DM m <sup>-3</sup>	167 <sup>a</sup>	157 <sup>a</sup>	275 <sup>a</sup>	273 <sup>a</sup>

\*The two years were analysed separately.

Therefore, comparison of the P load weights in 2004 and 2005 provided the possibility of predicting the influence of the crop. It can be assumed that an optimal wilting and preparation of

the swath makes a difference in machinery performance and, subsequently, in the total cost of silage production. The capacity in terms of tonne DM loaded, transported and unloaded per hour differed between two systems. The loading capacity expressed as quantity of forage DM loaded per hour was significantly higher in P system and resulted in a similar loading time as in TS 40/50 systems (Table 1). On the other hand, the density of loaded forage in TS40 wagon ( $67 \text{ kg DM m}^{-3}$ ) and TS 50 wagon ( $88 \text{ kg DM m}^{-3}$ ) were higher than in P wagons (2004 = 50 and 2005 =  $78 \text{ kg DM m}^{-3}$ ). Thus, precision chopping leads to an increase in loaded density. Differences in fuel consumption between the systems were marginal but a great variation was seen between the years. Both loading systems were found to have considerably lower fuel consumption in 2005 than in 2004. It is suggested that besides wagon volume, fuel consumption depended also on crop DM and transport distance. The importance of distance was similarly found by Frost and Binnie (2005) comparing a self-propelled forage harvester and a self-loading forage wagon.

Table 2. Distribution of particle length in forage from Pöttinger rotor cutter wagon and Taarup/Sahlström precision chopping wagons.

Harvest Wagon	2004		2005	
	Pöttinger	Taarup TS 40	Pöttinger	Taarup TS 50
Nominal, mm	34	25	34	25
Average, mm	58	25	56	35
Highest 25%, mm	99	38	57	39
Lowest 25%, mm	34	16	20	16

Two cutting and loading techniques resulted in different distributions of particle lengths (Table 2). Forage from the P system showed a longer particle length than forage in the TS systems in both years. It would be expected that forage from TS systems being packed to a higher density, but results showed a similar final density of the stored silage between systems (Table 1). Arvidsson and Lingvall (2005) also identified this tendency in a similar laboratory experiment. It was questioned whether less lacerated forage would influence the feed intake and animal performance. However, no significant differences were found between the two silages (Bertilsson, 2008).

## Conclusions

- The rotor-cutter loading system was significantly superior to the precision chopping in loading capacity and total ensiled forage DM per hour.
- No difference in silage fermentation or hygienic quality between the two loading systems were identified. All silages reached above the 'good' classification.
- The loading capacity is closely related to forage DM.
- The transport distance is an important factor influencing ensiling capacity.
- Fuel consumption was mainly related to forage DM and transport distances.

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## **$\alpha$ -tocopherol and $\beta$ -carotene in baled silage and haylage**

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### **Abstract**

The influence of forage conservation on content of  $\alpha$ -tocopherol and  $\beta$ -carotene in grass preserved as silage or haylage was investigated. Grass was baled in small square and small round bales as silage (SIL) containing 300 g dry matter (DM) kg<sup>-1</sup>, and two haylages; haylage 1 (HL1) containing 500 and haylage 2 (HL2) containing 600 g DM kg<sup>-1</sup>. Analysis of variance was carried out for effect of DM, bale type and interactions between DM and bale type. Pearson correlation coefficients were calculated between fermentation variables and  $\alpha$ -tocopherol or  $\beta$ -carotene. Results showed that interactions between DM and bale type were present and SIL in square bales contained the highest level of  $\alpha$ -tocopherol and  $\beta$ -carotene. Silage in square bales and HL2 in both round and square bales contained the highest proportion (0.6) of the initial concentration of  $\alpha$ -tocopherol in the fresh crop. The corresponding proportion for  $\beta$ -carotene was 0.9. Correlation coefficients between fermentation variables and  $\alpha$ -tocopherol or  $\beta$ -carotene were present but weak. No clear effect of DM level on vitamin content was present and other factors than DM level or conservation characteristics may be more relevant for preservation of vitamins in forage during conservation.

Keywords: silage, haylage, vitamin, bales

### **Introduction**

Feed-table data on vitamin content in forages is based mainly on studies of hay and silo-stored silage. However, wrapped forages such as baled silage and haylage are commonly used in Sweden, especially in equine rations. Specific investigations of factors that influence the content of  $\alpha$ -tocopherol and  $\beta$ -carotene in wrapped forages are scarce. As baled silage differs from silo-stored silage in fermentation characteristics even at the same dry matter (DM) content (Haigh, 1990; Field and Wilman, 1996; Fychan and Jones, 1996), the vitamin content might also be affected differently, considering the broad range of DM levels in wrapped forages, 210 to 810 g DM g kg<sup>-1</sup> (Holmquist & Müller, 2002). Therefore, an experiment investigating the content of  $\alpha$ -tocopherol and  $\beta$ -carotene in wrapped forages of three different DM levels and of two bale types (round and square) was conducted.

### **Materials and methods**

The crop used was a first cut from a permanent grass ley consisting of 0.45 timothy (*Phleum pratense*), 0.45 meadow fescue (*Festuca pratensis*) and 0.1 couch grass (*Agropyron repens*) grown and harvested in the middle of June in Uppsala, Sweden. Four hours after mowing, the crop had reached a DM level of 300 g kg<sup>-1</sup> and 1/3 of the cut crop was baled to produce silage (SIL). The remaining crop was tedded and left to wilt further, and 24 hours after mowing, haylage 1 (HL1) was baled containing 500 g DM kg<sup>-1</sup>. The last third of the crop was then tedded again and wilted to 600 g DM kg<sup>-1</sup> which was reached 34 hours after mowing, and baling of haylage 2 (HL2) took place. Baling was done with two different balers for each DM level, one producing small round bales (diameter 57 cm, height 68 cm) and one producing

small square bales (80 x 48 x 36 cm). All forages were conserved in long-stemmed state, and no additive was used. Bales were wrapped with ten layers of white, 360 mm wide and 25  $\mu$ m thick stretch film (0.5 overlap of layers, 1.7 pre-stretch of the film) within two hours after baling. In total, 36 bales were produced, with six replicates for each treatment combination. Sampling of the wilted material was done at three different places in the field immediately before baling, to produce one pooled sample per DM treatment. Sampling of conserved forages was done in the bales after eleven months of storage, using a stainless steel corer. Analysis comprised DM, pH, and content of ash, water soluble carbohydrates (WSC), crude protein, neutral detergent fibre (exclusive of ash), organic acids, alcohols,  $\alpha$ -tocopherol and  $\beta$ -carotene. All analytical methods and the experiment in detail were described by Müller *et al.* (2007). Analysis of variance was carried out for effects of DM, bale type and interactions between DM and bale type. Pearson correlation coefficients were calculated between fermentation variables and  $\alpha$ -tocopherol or  $\beta$ -carotene using SAS linear correlation procedure.

## Results and discussion

The chemical composition of the wilted crops is presented in Table 1. Interactions between DM and bale type were present for all variables except butyric acid content (Table 2). SIL in square bales contained more lactic acid,  $\alpha$ -tocopherol and  $\beta$ -carotene than SIL in round bales. Within HL1, the content of WSC was higher in square than in round bales. Within HL2, no differences between bale types were found.

Table 1. Composition ( $\text{g kg}^{-1}$  DM unless otherwise mentioned) of the wilted crops before baling of silage (SIL), haylage 1 (HL1) and haylage 2 (HL2) ( $n = 1$ ).

Variable	SIL	HL1	HL2
Dry matter, $\text{g kg}^{-1}$	319	433	596
Ash	69	72	64
Crude protein	127	145	110
Water soluble carbohydrates	125	118	141
Neutral detergent fibre	586	574	596
$\alpha$ -tocopherol, $\text{mg kg}^{-1}$ DM	74	68	53
$\beta$ -carotene, $\text{mg kg}^{-1}$ DM	50	53	31

Table 2. Composition ( $\text{g kg}^{-1}$  DM unless otherwise mentioned) of silage (SIL), haylage 1 (HL1) and haylage 2 (HL2) conserved in round and square bales ( $n = 6$ ).

Variable	SIL Round	SIL Square	HL1 Round	HL1 Square	HL2 Round	HL2 Square	SEM	P
Dry matter, $\text{g kg}^{-1}$	286.4 <sup>a</sup>	283.4 <sup>a</sup>	507.1 <sup>b</sup>	502.6 <sup>b</sup>	577.1 <sup>c</sup>	551.1 <sup>c</sup>	14.2	< 0.05
pH	5.23 <sup>a</sup>	5.08 <sup>a</sup>	5.58 <sup>b</sup>	5.43 <sup>b</sup>	5.47 <sup>b</sup>	5.55 <sup>b</sup>	0.06	< 0.05
Ash	84.4 <sup>a</sup>	88.5 <sup>a</sup>	79.1 <sup>a</sup>	80.4 <sup>a</sup>	75.8 <sup>b</sup>	74.3 <sup>b</sup>	2.87	< 0.05
Water soluble carbohydrates	11.9 <sup>a</sup>	8.1 <sup>a</sup>	47.4 <sup>b</sup>	65.3 <sup>c</sup>	55.3 <sup>b,c</sup>	65.6 <sup>c</sup>	5.12	< 0.05
Lactic acid	28.6 <sup>a</sup>	37.3 <sup>b</sup>	2.0 <sup>c</sup>	4.2 <sup>c</sup>	1.4 <sup>c</sup>	1.4 <sup>c</sup>	2.27	< 0.05
Acetic acid	14.3 <sup>a</sup>	13.0 <sup>a</sup>	2.4 <sup>b</sup>	2.8 <sup>b</sup>	2.1 <sup>b</sup>	1.9 <sup>b</sup>	0.88	< 0.001
2,3-butanediol	36.7 <sup>a</sup>	33.3 <sup>a</sup>	1.3 <sup>b</sup>	3.0 <sup>b</sup>	1.0 <sup>b</sup>	0.7 <sup>b</sup>	4.68	< 0.01
Ethanol	34.4 <sup>a</sup>	31.3 <sup>a</sup>	23.9 <sup>b</sup>	19.6 <sup>b,c</sup>	16.2 <sup>c</sup>	18.4 <sup>b,c</sup>	2.01	< 0.05
Butyric acid	1.6	2.9	Nd	0.3	Nd	0.7	1.49	> 0.24
$\alpha$ -tocopherol, $\text{mg kg}^{-1}$ DM	31.9 <sup>a,c</sup>	42.5 <sup>b</sup>	26.8 <sup>a,c</sup>	24.9 <sup>a</sup>	33.6 <sup>c</sup>	32.3 <sup>a,c</sup>	2.65	< 0.05
$\beta$ -carotene, $\text{mg kg}^{-1}$ DM	20.6 <sup>a,c</sup>	44.4 <sup>b</sup>	20.8 <sup>a,c</sup>	10.3 <sup>c</sup>	27.1 <sup>a</sup>	26.1 <sup>a</sup>	4.98	< 0.01
<sup>A</sup> Proportion of initial content								
$\alpha$ -tocopherol	0.43 <sup>a</sup>	0.57 <sup>b</sup>	0.39 <sup>a</sup>	0.36 <sup>a</sup>	0.63 <sup>b</sup>	0.61 <sup>b</sup>	-	< 0.05
$\beta$ -carotene	0.41 <sup>a</sup>	0.88 <sup>b</sup>	0.39 <sup>b</sup>	0.19 <sup>a</sup>	0.87 <sup>b</sup>	0.84 <sup>b</sup>	-	< 0.05

<sup>a, b, c</sup>Different superscripts within a row indicate significant difference. Nd, not detected.

<sup>A</sup>Proportion of  $\alpha$ -tocopherol and  $\beta$ -carotene of initial content in the wilted crop (Table 1).

No general effect of bale type was found on content of  $\alpha$ -tocopherol or  $\beta$ -carotene. A main effect of DM was present, as SIL differed from HL1 and HL2 in more variables than HL1 and HL2 differed from each other (Table 2). In general, HL1 contained less  $\alpha$ -tocopherol and  $\beta$ -carotene than SIL or HL2. The explanation for this result is uncertain, and may depend on other factors than the conservation characteristics, e.g. differences in weather during wilting, uneven swaths and field-variation in vitamin content of the crop (Ballet *et al.*, 2000). However, in the conserved forages, the proportion of  $\alpha$ -tocopherol and  $\beta$ -carotene of the initial content in the wilted crops was higher in SIL in square bales and HL2 in round and square bales compared to the other treatment combinations (Table 2). Pearson correlation coefficients between fermentation variables and  $\alpha$ -tocopherol or  $\beta$ -carotene were weak (Table 3). The highest correlation coefficients were found between  $\beta$ -carotene and lactic acid content (0.44) and  $\alpha$ -tocopherol and lactic acid content (0.41). This may imply that a lactic acid fermentation or presence of lactic acid can be beneficial for preservation of  $\alpha$ -tocopherol and  $\beta$ -carotene in wrapped forages.

Table 3. Pearson correlation coefficients between  $\alpha$ -tocopherol or  $\beta$ -carotene ( $\text{mg kg}^{-1}$  DM) and fermentation variables ( $\text{g kg}^{-1}$  DM).

Variable	$\alpha$ -tocopherol <sup>a</sup>	P - value <sup>b</sup>	$\beta$ -carotene <sup>a</sup>	P - value <sup>c</sup>
Dry matter	-0.34	0.04	-0.27	0.11
pH	-0.34	0.04	-0.40	0.02
Water soluble carbohydrates	-0.39	0.02	-0.35	0.04
Lactic acid	0.41	0.0	0.44	0.01
Acetic acid	0.34	0.04	0.33	0.05
2,3-butanediol	0.35	0.07	0.23	0.25
Ethanol	0.24	0.16	0.05	0.79
Butyric acid	0.35	0.19	0.20	0.46

<sup>a</sup>Pearson correlation coefficient. <sup>b</sup>Level of significance for correlations between  $\alpha$ -tocopherol and other variables

<sup>c</sup>Level of significance for correlations between  $\beta$ -carotene and other variables.

## Conclusions

The content of  $\alpha$ -tocopherol and  $\beta$ -carotene was higher in SIL in square bales than in any other treatment combination. SIL in square bales and HL2 in round and square bales retained more of the initial content of  $\alpha$ -tocopherol and  $\beta$ -carotene in the wilted crops than the other treatments. A weak positive correlation was found between  $\alpha$ -tocopherol and lactic acid as well as between  $\beta$ -carotene and lactic acid. Other factors apart from DM level and conservation characteristics may be more relevant for preserving the content of  $\alpha$ -tocopherol and  $\beta$ -carotene of fresh grass during conservation, and the area needs further research.

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## Storage of small bale silage and haylage

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### Abstract

Changes in fermentation variables and microbial composition during storage of small-bale silage and haylage were investigated. Grass from the same sward and cut was used to produce silage (SIL) with 350 g dry matter (DM) kg<sup>-1</sup>, haylage (HLL) with a lower DM content (550 g kg<sup>-1</sup>) and haylage (HLH) with a higher DM content (700 g kg<sup>-1</sup>), using a conventional hay baler. The bales were wrapped individually and stored for a short (2 months) or long (14 months) term before opening and sampling. Analysis of variance was carried out for effects of DM, storage period and interactions between DM and storage period. Pearson correlation coefficients were calculated between short- and long-term stored bales for all variables. The results showed that interactions between DM and storage times were present. SIL changed in fermentation variables during storage, while HLL and HLH remained almost unchanged. Within storage times, SIL had higher concentrations of fermentation products and a lower pH than either of the two haylage types. Linear correlations between short- and long-term stored bales were present for all chemical variables, but not for microbial variables. In conclusion, changes in chemical fermentation variables during storage of small bales can be expected in silage but to a much smaller extent in haylage.

Keywords: silage, haylage, bales, storage

### Introduction

During recent years, use of wrapped forages, such as silage and haylage, has increased in equine nutrition and has partly replaced hay as forage for horses. As more than 0.75 of the Swedish horse population is housed in stables with only one to four horses (Persson, 2005), conventional big-bale silage or haylage can be difficult to use due to the limited aerobic storage stability of opened bales and the low daily consumption. Smaller bales, which can be fed within a few days, are therefore of interest but machinery equipment adapted for production of small-bale silage and haylage is lacking and conventional hay-balers are often used, which may result in porous bales with a low level of compaction (Müller, 2005). Forage with a low compaction level has been shown to have higher oxygen diffusion rates (Rees *et al.*, 1983), and oxygen may enter the forage through damage in the stretch film or by diffusion through the plastic (Borreani and Tabacco, 2005). Also, horse-owners sometimes prefer a very high dry matter (DM) level in wrapped forages (Holmquist and Müller, 2002). As forage with high DM (above 500 g DM kg<sup>-1</sup>) also has more resistant stalks (Keller, 1998), higher porosity (Williams, 1994) and a larger residual content of water soluble carbohydrates (Müller, 2005) compared to ensiled forage with lower DM level, the combination of low compaction and high DM may result in greater risk of unwanted changes in biochemistry and microbiology in the forage during storage. With this background, an experiment investigating changes in fermentation variables and microbial composition during storage of small-bale silage and haylage, originating from the same crop but differing in DM levels, was therefore performed.

## Materials and methods

A first cut from a permanent grass ley, consisting of approximately 0.5 timothy (*Phleum pratense*), 0.4 meadow fescue (*Festuca pratensis*) and 0.1 couch grass (*Agropyron repens*), was used. The crop was cut with a mower conditioner with flails and then wilted to three DM levels: 350, 550 and 700 g kg<sup>-1</sup> DM to produce silage (SIL), haylage with a lower DM level (HLL) and haylage with a higher DM level (HLH). The fresh crop contained on average 103 g crude protein, 60 g ash and 615 g neutral detergent fibre kg<sup>-1</sup> DM at harvest. Baling was done using a conventional hay baler producing square bales of size 80 x 48 x 36 cm. Wrapping was done within 2 hours from baling with eight layers of white, 360 mm wide and 25 µm thick stretch film (0.5 overlap between layers and pre-stretching of the film by 1.7). The bales were stored in a fenced bale-yard, stacked in four layers on wooden pallets. Sampling of the wrapped bales was done after 2 (short term storage) and 14 (long term storage) months of storage. At both sampling occasions, 18 bales (6 bales per DM treatment) were sampled. Samples were analysed for DM, pH, water soluble carbohydrates, organic acids, alcohols, ammonia nitrogen, yeast, mould, enterobacteria and clostridial spores. At opening, the bale surface was inspected for visible fungal growth. All analytical methods, as well as the experiment in detail, have been described by Müller *et al.* (2007). Statistical evaluation included variance analysis of the effects of DM and storage time, interaction between DM and storage time and also linear correlation analysis of variables between storage times.

## Results and discussion

Table 1. Chemical (g kg<sup>-1</sup> DM) and microbial composition (CFU g<sup>-1</sup>) in silage (SIL) and haylages (HLL and HLH) stored for 2 (short term storage) or 14 (long term storage) months.

Variable	SIL short term storage	SIL long term storage	HLL short term storage	HLL long term storage	HLH short term storage	HLH long term storage	sem	P DM level x storage time
Dry matter g kg <sup>-1</sup>	303	309	577	552	684	659	10.6	ns
pH	4.88 <sup>a</sup>	4.50 <sup>b</sup>	5.62 <sup>c</sup>	5.48 <sup>c</sup>	5.82 <sup>c</sup>	5.66 <sup>c</sup>	1.4x10 <sup>-6</sup>	< 0.0001
Lactic acid	33.5 <sup>a</sup>	43.0 <sup>b</sup>	2.6 <sup>c</sup>	1.3 <sup>c</sup>	0.3 <sup>c</sup>	0.4 <sup>c</sup>	0.89	< 0.0001
Acetic acid	6.6 <sup>a</sup>	15.9 <sup>b</sup>	1.4 <sup>c,d</sup>	2.6 <sup>d</sup>	0.6 <sup>c</sup>	0.8 <sup>c</sup>	0.64	< 0.0001
Butyric acid	0.8	0.7	< 0.01	< 0.01	< 0.01	< 0.01	0.09	ns
Succinic acid	12.5 <sup>a</sup>	10.8 <sup>b</sup>	2.0 <sup>c,d</sup>	2.6 <sup>c</sup>	1.3 <sup>c,d</sup>	2.0 <sup>c,d</sup>	0.44	< 0.01
Ethanol	23.5	30.4	7.9	19.8	4.3	13.0	1.71	ns
2,3-butanediol	21.2 <sup>a</sup>	16.5 <sup>b</sup>	0.9 <sup>c</sup>	< 0.01	< 0.01	< 0.01	0.75	< 0.01
Water soluble carbohydrates	26	5	69	44	71	50	3.8	ns
Ammonia-N g kg <sup>-1</sup> total N	105.1	118.6	29.0	34.1	14.6	18.0	3.72	ns
Yeast <sup>B</sup>	4.5	2.2	5.5	6.9	5.2	6.7	1.93x10 <sup>6</sup>	ns
Mould <sup>B</sup>	< 1.7	2.9	< 1.7	< 1.7	< 1.7	< 1.7	2.26x10 <sup>2</sup>	ns
Enterobacteria <sup>B</sup>	< 1.7	< 1.7	2.8	< 1.7	< 1.7	< 1.7	2.05x10 <sup>2</sup>	ns
Clostridial spores <sup>B</sup>	5.1	4.4	2.5	< 1.7	1.7	< 1.7	5.79x10 <sup>4</sup>	ns

ns, not significant.

<sup>a, b, c, d</sup> Different superscript letters indicate differences among means within row at the P-level presented.

<sup>A</sup> pH-values are means of H<sup>+</sup> transformed to pH, while sem-values are H<sup>+</sup>.

<sup>B</sup> CFU-values are CFU g<sup>-1</sup> transformed to <sup>10</sup>log-values, while SEM-values are CFU g<sup>-1</sup>.

General effects of DM level and of storage time were found, but interactions between DM level and storage time were also present (Table 1) and revealed that effect of storage time were present in treatment SIL only. In SIL, the fermentation process seemed to continue during storage, as lactic and acetic acid increased and pH decreased. In HLL and HLH, no



effect of storage time was detected in any variable. Within both storage times, SIL differed from HLL and HLH in chemical composition, but the two haylages did not differ from each other, except from acetic acid content which was somewhat higher in HLL than in HLH within long-term storage. Microbial variables did not differ among any treatment combinations. No surface fungi were detected.

Pearson correlation coefficients for chemical variables between short- and long-term storage are presented in Table 2. Correlation coefficients were high for several chemical variables (Table 2), but no statistically significant linear correlation between short and long term storage were found for microbial variables (data not shown).

Table 2. Linear correlation analysis (Pearson correlation coefficients) for chemical composition between short-term and long-term stored silage and haylage.

Variable	Pearson correlation coefficient	P
Dry matter	0.97	< 0.0001
pH	0.98	< 0.0001
Lactic acid	0.99	< 0.0001
Acetic acid	0.97	< 0.0001
Butyric acid	0.69	< 0.01
Succinic acid	0.97	< 0.0001
Ethanol	0.82	< 0.0001
2,3-butanediol	0.97	< 0.0001
Water soluble carbohydrates	0.85	< 0.0001
Ammonia-N	0.89	< 0.0001

## Conclusions

The dry matter level of wrapped forages interacted with storage time. Silage (SIL) changed in fermentation variables during storage time, but haylages did not change to the same magnitude. Chemical variables were correlated between short- and long-term stored bales, while no statistically significant linear correlation coefficients were found for microbial variables. In conclusion, changes in chemical fermentation variables during storage can be expected to a higher degree for silage than for haylage. This also means that samples taken from haylage bales after 2 months of storage can also be representative of chemical composition of the haylage after 14 months of storage, though this may not be true for silage.

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# Effects of slurry application and wide-spreading of forage on silage quality

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## Abstract

Windrowing swaths and manuring leys have been suspected to contaminate the grass with harmful microbes, which can contaminate silage and milk. We studied how silage quality was affected by wide-spreading the grass at mowing followed by windrowing (WS + W, 2002-2004), broadcast neat slurry 46 m<sup>3</sup> ha<sup>-1</sup> in spring or after 1st cut (2003-2004), and additive (2004) in trials using 1st and 2nd cut grass, which was baled and wrapped. The microbiological quality was analysed regarding clostridia and aerobic bacterial spores, enterococci, mould, and yeast. WS + W did not impair microbiological quality to any practical extent in any cut, not even in combination with slurry. Slurry raised the clostridia and aerobic bacterial spore counts to harmful levels in all cuts, when spread to the harvest in question. Slurry spread in spring still had a harmful effect in the second cut 2003. Additive (formic acid 430, ammonium formate 300, propionic acid 100, and benzoic acid 20 g kg<sup>-1</sup>), application 4.0-4.5 L Mg<sup>-1</sup>, had a small effect on microbiological quality, but improved fermentation quality.

## Introduction

Traditionally it has been recommended not to windrow swaths intended for silage because of risk of contamination with harmful soil microbes, which can contaminate silage and milk. However, windrowing is required to utilize the capacity of large choppers and loader wagons, which are becoming more common. Moreover, wilting is faster when setting the mower for spreading the grass over its full working width, instead of mowing it to windrows. Then windrowing is required before the wide-spread forage can be harvested. Wide-spreading and the subsequent windrowing also cause wheeling over the swaths, which is another potential risk of contamination. Another hygienic risk is spreading of manure on leys. This is becoming more common with farms expanding their herds so that all their land is turned to leys. Clostridia spores from soil and manure can cause bad silage fermentation, and spores can contaminate milk causing bad fermentation in cheese (Vissers *et al.*, 2007). The main aim of this project was to study how wide-spreading the grass at mowing followed by windrowing combined with broadcasting of liquid manure affects microbial and chemical quality of silage.

## Materials and methods

In all years 2002-2004, two cuts of mixed *Phleum pratense* and *Festuca pratensis* were harvested as wrapped round bales. In 2002 the only trial factor was wide-spreading + windrowing (WS + W) versus swathing without windrowing (later 'swathing'). In 2003-2004 slurry was added as a factor, and in 2004 also additive containing formic acid 430, ammonium formate 300, propionic acid 100, and benzoic acid 20 g kg<sup>-1</sup>. The average application at 1st cut was 4.5 and in 2nd cut 4.0 L Mg<sup>-1</sup>. The slurry treatments: 1) artificial NPK for both cuts, 2) slurry in spring and artificial NPK after 1st cut, and 3) artificial NPK in spring and slurry after 1st cut. The slurry was untreated neat slurry, dry matter 70 g kg<sup>-1</sup>, applied 46 m<sup>3</sup> ha<sup>-1</sup> both years. It contained (log<sub>10</sub>/g 2003/2004): 6.0/5.0 clostridia spores, 5.5/6.6 aerobic bacterial spores, 7.0/5.0 enterococci, 3.4/3.5 mould, and 2.0/2.0 yeast. It was completed with artificial NPK so that all treatments received a total of 90 kg ha<sup>-1</sup> soluble N. Spring spreading was



22-23.5.03 and 18.5.04. The 2nd spreading was 8-9 days after 1st cut 2003. In 2004 it was not until 35 days after 1st cut due to frequent rains and soft ground. The grass was shortest at the application after 1st cut 2003, then regrowth had barely started. At the other applications, the height was 21-26 cm. Both swaths and windrows were about 1.2 m wide. Swaths were about 0.25 m high before baling, windrows about 0.50 m. The windrower had one rotor with vertical axle (Krone KS 4.61/13). The tines were set 4 cm from the ground. Samples were taken of the herbage at mowing, just before baling, and from the bales in February-March. The samples before baling represented averages of the swath/windrow cross-sections; the variation in dry matter between top and bottom layers was not studied. The standard deviation of dry matter content in the different cuts before baling was 1.4-4.7 percentage points in the swaths, and 1.8-4.6 in the windrows.

Table 1. Treatments. + = yes, - = no (ws+w: - = swathing), 1 = in spring, 2 = after 1st cut.

Year-cut	02 - 1		02 - 2		03 - 1				03 - 2					
WS + W	-	+	-	+	-	+	-	+	-	+	-	+	-	+
Slurry					-	-	1	1	-	-	1	1	2	2
Additive														
n	4	4	4	4	11	11	8	8	6	6	6	6	8	6

Year-cut	04 - 1								04 - 2							
WS + W	-	-	+	+	-	-	+		-	-	+	+	-	+	-	+
Slurry	-	-	-	-	1	1	1		-	-	-	-	1	1	2	2
Additive	-	+	-	+	-	+	-		+	-	+	+	+	+	-	+
n	4	8	8	3	4	4	8		4	4	4	4	4	4	4	4

## Results and discussion

There were few significant interactions between the factors regarding microbial quality, and those found were small without practical relevance. In 2004, the interactions could be tested completely only for swaths/windrows. For the silage that year, all interactions could not be tested, because the trial was not completely factorial regarding additive. For clostridia and aerobic bacterial spores a recommendation has been max. log 3 spores g<sup>-1</sup> of silage (Spöndly 2003). The clostridia spore content in the silage was significantly higher with slurry in all four cuts, and in three cuts above the recommended level, when the slurry had been spread for the harvest in question (Figure 1A). The results were the same already in the swaths/windrows. The increase was roughly hundredfold (log 2). Sundberg (2002) suggests that if the increase in microbial count in the windrowed swath is in the region of tenfold, it can be taken as a proof of contamination. The effect of slurry on aerobic bacterial spores was similar (Figure 1B). In the two 2nd cuts the counts were above the recommended level also without slurry, but the increase with slurry was over tenfold, so the cause can be considered to be the slurry. The results were much the same already in the swaths/windrows. A large part of the bales from all cuts which had slurry for the harvest in question contained visible residues of slurry. In 2003, slurry in spring raised the clostridia spore count even in second-cut silage, *ca.* hundredfold ( $P < 0.001$ ). In 2004, slurry in spring had no considerable effect on second-cut silage, raising aerobic bacteria spore count with log 0.6 ( $P < 0.01$ ). The effect of slurry on enterococci was not as clear as on clostridia and aerobic spores. In all cuts except 1st cut 2003, there were plenty of enterococci already without slurry, roughly log 4–log 6, and slurry did not raise the levels. In 1st cut 2003 slurry raised the level in silage tenfold ( $P < 0.001$ ). Slurry did not increase the content of mould and yeast in the silage of any cut. Slurry for the harvest in question decreased crude protein content about 2.5 percentage points in both cuts 2003. It decreased ammonia levels 2-4 percentage points in three cuts. Otherwise slurry had small effect on chemical quality in all cuts. WS + W did not impair microbiological quality to any practical extent in any cut, not even in combination with slurry. It raised the aerobic bacteria spore count in the silage significantly in just one of six cuts, 2nd cut 2004, and then only threefold (Figure 2B). In 2nd cut 2003, WS + W increased the clostridia spores in the windrows ( $P < 0.01$ ) and the mould in the silage ( $P < 0.01$ ). However, the increases were

small, to the double, and the levels with WS + W were still low, log 1.25 and log 1.62, respectively. Finally, WS + W increased enterococci ( $P < 0.001$ ) in 1st cut 2004, but only threefold. The clostridia spore count was even lower with WS + W in the silage of 1st cut 2004, under a tenth compared to swathing. The level was too high with swathing (Figure 2A). The effect of additive on microbiological quality was rather small. In 1st cut it decreased clostridia spores from log 3.4 to log 2.9 ( $P < 0.01$ ). In 2nd cut it decreased enterococci from log 5.9 to 5.6 ( $P < 0.05$ ). The enterococci count was still high after that. Fermentation quality, on the other hand, was clearly improved by the additive. It decreased ammonia ( $P < 0.001$ ) and butyric acid ( $P < 0.01$ ) in both cuts. The pH without/with additive was 4.81/4.57 in 1st and 4.93/4.80 in 2nd cut.

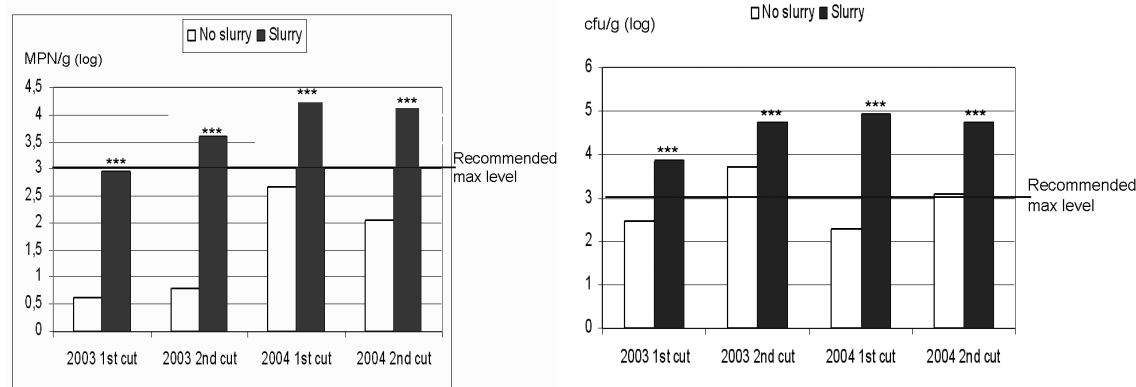


Figure 1. A) Clostridia spores in silage, averaged over wilting and additive treatments (MPN = most probable number). B) Aerobic bacterial spores in silage, averaged over wilting and additive treatments (cfu = colony forming units). \*\*\*  $P < 0.001$ .

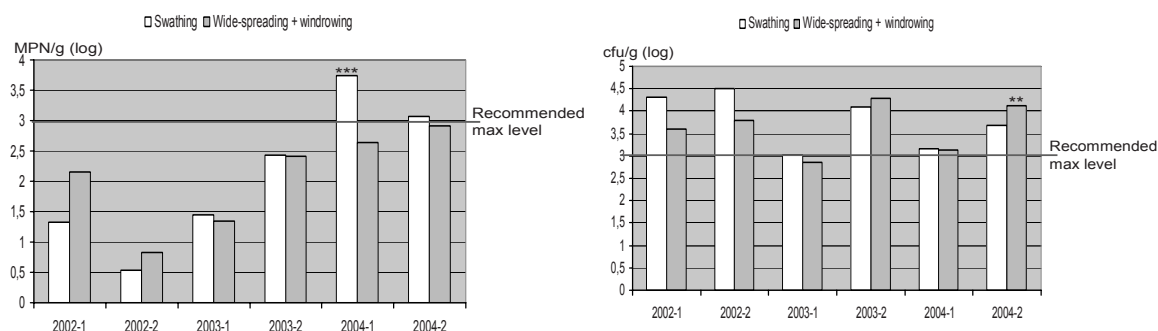


Figure 2. A) Clostridia spores in silage, averaged over fertilizer and additive treatments. \*\*\*  $P < 0.001$ . B) Aerobic bacterial spores in silage, average of fertilizer and additive treatments. \*\*  $P < 0.01$ .

## Conclusions

When using a high level of slurry at one occasion and waiting with the application for at least eight days after first harvest, the slurry raised the clostridia and aerobic bacterial spore counts to harmful levels in all cuts, when spread to the harvest in question. Slurry spread in spring still had a harmful effect in second cut 2003. WS + W did not impair microbiological quality to any practical extent in any cut, not even in combination with slurry. Additive had a rather small effect on microbiological quality, but improved fermentation quality.

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## Inoculation of experimental silages with different *Clostridium* spores

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### Abstract

Spore-forming *Clostridium* bacteria are one of the most common micro-organisms in wet silages that compromise forage quality and eventually contaminate dairy products. To study the effect of different *Clostridium* strains on silage quality, we inoculated 4 grass-dominated crops (mainly *Phleum pratense*, *Festuca pratensis*, *Trifolium pratense*) with one of 10 spore cocktails. Each cocktail contained between 1-3 different *Clostridium* strains (24 strains in total). The objective was to compare the clostridial activity among contaminated and control silages. 4 treatments (2 cuts x 2 DM levels) were inoculated with approximately 1000 spores g<sup>-1</sup> and ensiled in small laboratory silos. Results are shown from the later cut only. When analysed 3 months later, intensive clostridial fermentation had taken place in wet silages (approx. 300 g kg<sup>-1</sup> DM). While clostridial activity was lower, the differences among spore cocktails were larger in dry silages (approx. 400 g kg<sup>-1</sup> DM). Spore counts in wet silages were higher with cocktail 1 compared with other cocktails except cocktail 4. In drier silages, differences among applications were not as large and cocktail 1 produced similar spore counts as cocktail 5, 8, 9, 10 and the control. These results indicate that our previously selected *Cl. tyrobutyricum* strain (cocktail 1) is a suitable challenge organism for ensiling experiments that focus on the inhibition of clostridial activity.

Keywords: silage, fermentation, *Clostridium*, inoculation, contamination, spore count

### Introduction

The most efficient methods of avoiding spore-related problems in milk products are ones that stop the proliferation of clostridial spores in silage (Malmqvist and Spörndly 1993). Since 1989, we have tested the effect of different factors (e.g. additives, wilting methods, air leakage) on clostridial development in grass-based silages. In these experiments, we used a special strain of *Clostridium tyrobutyricum* (cocktail 1 in Table 1), which was isolated from blowing cheese by the Swedish Dairy Association in the mid-1980s. Over the years, we became increasingly concerned about the representativeness of this strain for Swedish farmyard silage. A research fund from the Swedish Farmers' Foundation for Agricultural Research ([www.lantbruksforskning.se](http://www.lantbruksforskning.se)) made it possible to investigate this question. The objective of this experiment was to compare the clostridial activity of laboratory-scale silages contaminated with various species and strains of *Clostridium* spores. Strains were selected from Swedish dairy products or farm-tank milk samples. Our intention was to find out how well our previously used strain competed in grass silage in comparison with the other strains.

### Materials and methods

A ley composed of approx. 65% temperate grasses (mainly *Phleum pratense*, *Festuca pratensis*) and 35% red clover (*Trifolium pratense*) was harvested in Uppsala, Sweden, on June 16 and at the end of the growing season (September 11). On both occasions, the fresh forage was wilted in the field to approx. 300 (low DM) and 400 g kg<sup>-1</sup> DM (high DM) before

being chopped in a stationary forage cutter. These 4 treatments (2 cuts x 2 DM levels) were each homogenized and 11 portions of 3 kg forage from each crop and DM level were then weighed into 125-l plastic bags. The spore suspensions listed in Table 1 (cocktails 1-10) were applied to the bags with spray bottles, adding approx.  $10^3$  spores  $g^{-1}$ . The eleventh portion was left untreated and served as a control. After mixing, triplicate silos (volume 1700 ml) were filled from each bag. Silos were stored at room temperature (19-22°C) for 103 to 126 days before they were opened, sampled and analysed for the parameters shown in Table 3.

Table 1. Spore cocktails used to inoculate our forage crops.

Cocktail No.	<i>Clostridium</i> spp.	Strain ID
1	<i>Cl. tyrobutyricum</i>	213
2	<i>Cl. tyrobutyricum</i>	216, 231, 366
3	<i>Cl. tyrobutyricum</i>	223, 352, 360
4	<i>Cl. tyrobutyricum</i>	194, 210, 263
5	<i>Cl. tyrobutyricum</i>	186, 362, 368
6	<i>Cl. tyrobutyricum</i>	192, 221, 365
7	<i>Cl. butyricum</i>	196, 203, 219
8	<i>Cl. bifermentans</i>	217, 233
9	<i>Cl. sporogenes</i>	199, 234
10	<i>Cl. sticklandi</i>	195

Source: Collection of the Swedish Dairy Association.

Before application to the herbage, the clostridial strains were multiplied on reinforced clostridial agar (Merck 1.05410) supplemented with 200 ppm D-cycloserine (Sigma C-6880) according to Jonsson (1990). After 7 days of cultivation the agar plate colonies were rinsed off with quarter-strength Ringer solution (Merck 1.15525). After centrifuging and washing, the suspensions were stored at +2 °C until used. Spore count in suspensions was determined by two different methods: a) microscopic counts in a counting chamber for first-cut forages and, as above, b) viable counts after cultivation on clostridial agar for third-cut forages. Spore cocktails were composed by randomly allocating up to 3 strains of one species per cocktail. Only cocktail 1 contained just our previously used strain. Silage data were analysed by analysis of variance for each cut and DM level separately because of different counting methods used in cocktails applied to first- and third-cut forages. Cocktail was the only class variable. Tukey's t-test was used to identify significant differences ( $P < 0.05$ ) among cocktails.

## Results and discussion

Viable LAB counts were low and clostridial counts were below detection level ( $< 50$  cfu  $g^{-1}$ ) in fresh forages (Table 2). Spore density of the suspensions varied in microscopic counts between log 7.1-9.8  $ml^{-1}$  and log cfu 2.0-7.3  $ml^{-1}$  in viable counts. The largest difference between the two counting methods was detected in *Cl. tyrobutyricum* strain #365 (7.4 log units) and the smallest in *Cl. sporogenes* strain #234 (0.6 log units). The average difference for all 24 strains was 3.4 log units. Microscopic counts are expected to overestimate the true spore count because not all visible spores will germinate.

Table 2. Forage composition from 2 cuts and 2 DM levels. CP – crude protein, WSC – water-soluble carbohydrates, BC - buffering capacity, LAB - lactic acid bacteria, na- not analysed.

Cut	DM level	DM g/kg	Water activity	Ash g/kgDM	CP g/kgDM	WSC g/kgDM	Nitrate g/kgDM	BC gLA/100gDM	LAB log cfu/g	<i>Clostridium</i> log cfu/g
1	Low	286	0.99	95	132	99	0.8	6.1	na	< 1.7
1	High	423	0.98	92	130	94	0.5	6.3	na	< 1.7
3	Low	299	0.98	122	190	103	0.8	6.6	2.6	< 1.7
3	High	408	0.97	122	191	97	0.5	6.6	3.3	< 1.7

Despite similar DM and WSC values, silages from the first and third cut differed in clostridial activity (values from first cut not shown). High DM silages from the first cut were free from butyric acid and contained low viable spore counts (log cfu 0.9-2.7 g<sup>-1</sup>). Differences between cocktails were marginal. The low DM crop from the first cut produced silages with very high spore counts (log 6.2-6.6 g<sup>-1</sup>), high ammonia-N values (168-263 g kg<sup>-1</sup> N) and high concentrations of butyric acid (23-32 g kg<sup>-1</sup> DM). Untreated control silages were only slightly better than inoculated silages. The lack of response to the spore inoculation was probably the result of the low amount of applied spores as spore dosage was based on microscopic counts. The low DM crop from the third cut produced silages with lower clostridial activity but with larger differences among treatments (Table 3). Silages inoculated with cocktail 1, 3 or 4 produced comparatively high spore counts, high butyric acid concentrations and high pH values. In the drier silages from the same cut, the application of cocktail 1, 5 or 9 produced the highest spore counts. However, only silages inoculated with cocktail 1 had in addition high butyric acid values.

Table 3. Composition of silages from the third cut. Means of triplicate silos.

Cocktail #	DM g/kg	pH	NH <sub>3</sub> -N g/kgN	Lactic acid g/kgDM	Acetic acid g/kgDM	Butyric acid g/kgDM	<i>Clostridium</i> log cfu/g
Low DM level:							
Control	300 <sup>ab</sup>	4.85 <sup>a</sup>	122	48 <sup>ab</sup>	11 <sup>c</sup>	7 <sup>d</sup>	2.7 <sup>a</sup>
1	289 <sup>c</sup>	5.19 <sup>b</sup>	128	15 <sup>e</sup>	12 <sup>c</sup>	27 <sup>a</sup>	5.7 <sup>b</sup>
2	299 <sup>ab</sup>	4.82 <sup>a</sup>	115	45 <sup>abc</sup>	11 <sup>c</sup>	9 <sup>cd</sup>	3.9 <sup>cd</sup>
3	296 <sup>bde</sup>	5.19 <sup>b</sup>	124	19 <sup>de</sup>	11 <sup>c</sup>	24 <sup>b</sup>	4.7 <sup>ce</sup>
4	291 <sup>cd</sup>	5.16 <sup>b</sup>	142	22 <sup>de</sup>	13 <sup>bc</sup>	24 <sup>b</sup>	4.9 <sup>bc</sup>
5	296 <sup>bde</sup>	5.05 <sup>bc</sup>	135	26 <sup>d</sup>	12 <sup>c</sup>	21 <sup>b</sup>	4.0 <sup>cd</sup>
6	296 <sup>bde</sup>	4.85 <sup>a</sup>	136	45 <sup>abc</sup>	16 <sup>a</sup>	10 <sup>cd</sup>	3.6 <sup>d</sup>
7	299 <sup>ab</sup>	4.94 <sup>ac</sup>	128	40 <sup>bc</sup>	13 <sup>bc</sup>	10 <sup>cd</sup>	3.3 <sup>ad</sup>
8	299 <sup>ab</sup>	4.88 <sup>a</sup>	111	43 <sup>abc</sup>	11 <sup>c</sup>	9 <sup>cd</sup>	3.2 <sup>ad</sup>
9	293 <sup>cde</sup>	5.04 <sup>bc</sup>	130	36 <sup>c</sup>	15 <sup>ab</sup>	11 <sup>c</sup>	3.7 <sup>d</sup>
10	301 <sup>a</sup>	4.82 <sup>a</sup>	140	50 <sup>a</sup>	13 <sup>bc</sup>	9 <sup>cd</sup>	3.3 <sup>ad</sup>
High DM level:							
Control	394 <sup>a</sup>	5.03 <sup>bcd</sup>	87	35 <sup>bc</sup>	6 <sup>c</sup>	< 0.4 <sup>a</sup>	2.6 <sup>bcd</sup>
1	391 <sup>ab</sup>	5.11 <sup>a</sup>	84	26 <sup>e</sup>	3 <sup>e</sup>	13 <sup>b</sup>	3.8 <sup>ab</sup>
2	391 <sup>ab</sup>	4.94 <sup>ef</sup>	84	38 <sup>ab</sup>	7 <sup>a</sup>	2 <sup>a</sup>	2.4 <sup>cd</sup>
3	395 <sup>a</sup>	5.02 <sup>bcd</sup>	81	32 <sup>cd</sup>	5 <sup>cd</sup>	6 <sup>c</sup>	2.0 <sup>cde</sup>
4	392 <sup>ab</sup>	5.06 <sup>abc</sup>	86	33 <sup>cd</sup>	5 <sup>cd</sup>	4 <sup>c</sup>	1.8 <sup>de</sup>
5	390 <sup>b</sup>	5.08 <sup>ab</sup>	83	31 <sup>d</sup>	5 <sup>cd</sup>	9 <sup>d</sup>	4.1 <sup>a</sup>
6	394 <sup>a</sup>	4.97 <sup>def</sup>	81	37 <sup>ab</sup>	7 <sup>ab</sup>	1 <sup>a</sup>	<1.7 <sup>e</sup>
7	393 <sup>ab</sup>	4.94 <sup>ef</sup>	85	39 <sup>a</sup>	7 <sup>a</sup>	< 0.4 <sup>a</sup>	2.3 <sup>cd</sup>
8	391 <sup>ab</sup>	5.01 <sup>bcd</sup>	85	37 <sup>ab</sup>	6 <sup>bc</sup>	< 0.4 <sup>a</sup>	3.2 <sup>abc</sup>
9	393 <sup>ab</sup>	4.93 <sup>f</sup>	83	39 <sup>a</sup>	7 <sup>ab</sup>	< 0.4 <sup>a</sup>	4.0 <sup>a</sup>
10	394 <sup>a</sup>	5.00 <sup>cde</sup>	78	36 <sup>ab</sup>	7 <sup>ab</sup>	< 0.4 <sup>a</sup>	2.6 <sup>bcd</sup>

## Conclusions

Inoculation of grass-based forage with our previously used *Clostridium tyrobutyricum* strain (cocktail 1) resulted in silages with higher or at least as high clostridial activity as the other spore cocktails. This indicates that this particular strain is a suitable challenge organism for experiments focusing on the inhibition of clostridial activity in silages.

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## Handling round bale silage after stretch-film application

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### Abstract

The aim of this study was to investigate whether or not there were any differences in plastic seal integrity or round bale silage quality depending on the time between stretch-film application and handling. 70 bales were rolled and wrapped on two occasions with a Vicon 1601 Kombibaler. The first cut was a ley with a clover/grass dry matter (DM) proportion of 40/60 (420 g DM kg<sup>-1</sup>) and the second cut was pure grass (750 g DM kg<sup>-1</sup>). Handling of the bales was made at 5 different times after baling: within 1 h, at approximately 3 h, at 24 h, after 3 days and after 10 days. A Trima Quadrigrip was used with enough gripping force to move the bales without them slipping. There was also one treatment where the bales were not handled at all. Seal integrity was measured twice: after 6 weeks and after at least 13 weeks. At the later occasion, the bales were unwrapped, examined for yeast and mould and chemically analysed. The result shows no clear evidence that any time would be better or worse to handle round bales within the first 10 days after stretch-film application.

Keywords: bale handling, tightness

### Introduction

It is more and more common to produce round bale silage in combined machines that produce the bale and apply the stretch-film. That leaves the wrapped bales in the field. In Sweden, the bales are usually transported on a wagon to the farm for storage under bird nets. The loading and unloading of the wagon is done by a bale grip mounted on the front loader of the tractor. This raises the question: ‘how to handle the bales without damaging the integrity of the plastic seal?’ There is a widespread belief among farmers and contractors that the bales must be handled at a certain time, either within the first few hours after wrapping before film lamination takes place, or a week later when film lamination is completed and no more gas is produced in the bale. However, these ideas have yet to be fully investigated. Randby and Fyhri (2005) presented inconsistent results when comparing bales moved immediately versus after 4-6 days using different equipment. The intention of the present experiment is to investigate the effect of time between wrapping and moving on the integrity of the plastic seal on the bales.

### Material and methods

The present study took place in southern Sweden and comprised 70 round bales from two cuts. The bales were made from a clover/grass crop of proportions 40/60 on a dry matter (DM) basis in the first cut, and a grass-only crop in the second cut. The crop was pre-wilted to 420 g DM kg<sup>-1</sup> in the first cut and to 750 g DM kg<sup>-1</sup> in the second cut. No silage additives were used. The bales were pressed and wrapped in the same machine, a Vicon RV 1601 Combibale (flex chamber) adjusted to produce bales with a diameter of 120 cm. The wrapper was calibrated to apply 6 layers of film with 50% overlapping and 70% stretching. The

stretch-film (Tenospin 750 x 0.0025 x 1500 m) was supplied by Trioplast AB and was 0.0025 mm thick. The 5 treatments (6 bales per treatment) differed according to when the bales were handled using a Trima Quadrogrip after wrapping: (1) within 1 hour, (2) between 3 and 5 hours, (3) at 24 hours, (4) after 3 days and (5) after 10 days. Five bales in a control treatment were not handled at all. At each handling, the bales were lifted twice and moved with the gripper, while applying a hydraulic pressure of 100 bar and resulting in a force on the bale of 14,500 N, measured on a scale mounted between the gripping arms. The tractor with bales in the gripper passed wooden logs on the ground, simulating the stress on the bales that occur when loaded on a wagon in the field.

Measurements of bale seal integrity were made on two occasions. The first at 6 weeks after wrapping and the second just before opening, which occurred 19 weeks after wrapping for bales harvested in the first cut and 13 weeks after wrapping in the second cut. Seal integrity was described as the time for an applied pressure of -200 Pa (vacuum) to rise to -150 Pa. The pressure was produced by a pump and a valve prior to measurement as described by Jonsson (2001). As a complement to the seal integrity measurement, the area of visible yeast and mould on the silage surface was measured when the bales were opened. To compare treatments, the GLM procedure in SAS (2002) was used to present least square means with cut and handling time as class variables.

## Results

The mean bale density was 158 kg DM kg<sup>-1</sup> in the first cut and 180 in the second. Silage quality was good for both cuts. No bales were found to have obvious inferior hygienic quality. The mean seal integrity of the bales in cut 1 and cut 2 are illustrated in Table 1. Seal integrity 1 was measured 6 weeks after ensiling. Seal integrity 2 and the prevalence of both yeast and mould visible on the surface were measured when the bales were opened.

Table 1. Mean seal integrity and prevalence of yeast and mould among round bales from two cuts.

	DM	pH	Ammonia-N, g kg <sup>-1</sup> tot N	Seal integrity 1, Seconds	Seal integrity 2, Seconds	Yeast, % of bale area	Mould, % of bale area
Cut 1	42.3	4.7	99	1144 *	541 *	0.01	0.17
Cut 2	75.7	5.6	16	1312 *	662 *	0.05	0.65
Sign.	< 0.0001	< 0.0001	< 0.001	ns	ns	ns	< 0.01

Numbers with \* differ significantly from the value 100 seconds,  $P < 0.05$ .

Table 2. Seal integrity and prevalence of yeast and mould on bales handled differently. Differences are tested using Least Square Means of 2 cuts.

Treatment	Seal integrity 1, Seconds	Seal integrity 2, Seconds	Yeast %	Mould %
Not handled	1333 *	354ab	0.15a	0.64
1 h	1212 *	606ac *	0.00b	0.39
3-5 h	1493 *	986c *	0.01ab	0.46
24 h	1323 *	611ac *	0.00b	0.24
3 days	1238 *	650ac *	0.00b	0.15
10 days	769 *	402ab	0.00b	0.54

Numbers with \* differ significantly from the value 100 seconds,  $P < 0.05$ .

Numbers in the same column with no letter in common differ significantly at  $P < 0.05$ .

There were no differences in seal integrity between cuts, but more surface mould was visible in the second cut. Average seal integrity was significantly above the limit of 100 seconds in both cuts. Periods of 100 seconds, or sometimes 60 seconds, are used to determine a basic level of seal integrity, often classified as sufficient under practical conditions (Jonsson, 2001).



Table 2 and Figure 1 present the mean seal integrity and prevalence of yeast and mould on bales at each handling occasion averaged over both cuts. Generally, neither cut nor handling time significantly affected the dependent variables presented in Tables 1 or 2. However, an analysis of contrasts reveals that certain treatments do affect seal integrity 2 and the prevalence of yeast. The bales handled 3-5 hours after wrapping had a better seal than those handled either after 10 days, or not handled at all. Furthermore, bale seal integrity was not significantly over 100 seconds for these latter handling times.

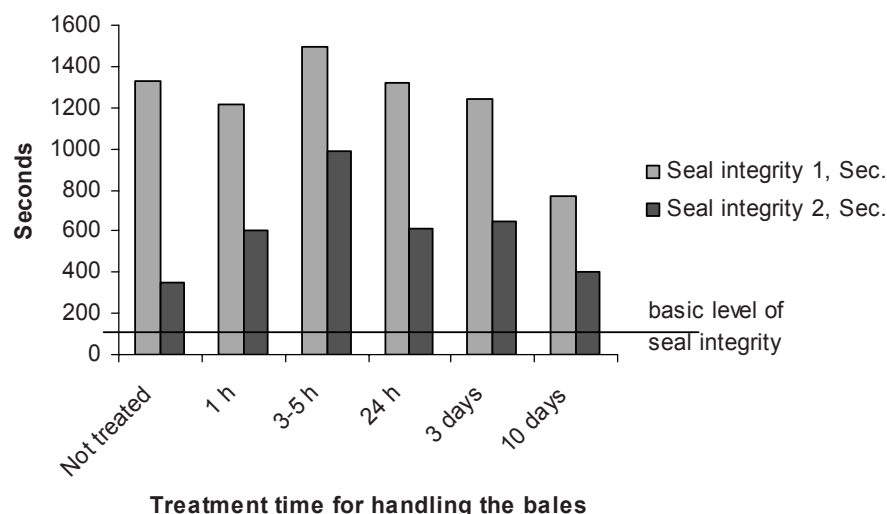


Figure 1. Illustration of the seal integrity of round bales handled at different times after wrapping. Seal integrity measured in seconds to increase a pressure from -200 Pa to -150 Pa (vacuum).

## Conclusion and recommendation

The belief that bales are more sensitive to handling between 3 and 24 hours after wrapping is unfounded. The bales with the lowest seal integrity, as well as the few bales where the film split during handling, were all found outside this time range. Bales which were not handled at all, and bales handled at day 10, had poorer seal integrity than those handled at 3-5 h.

Observations from this project also highlight the importance of the power used for gripping. Using too high a power causes the bales to crack. The appropriate power certainly depends on the type of bale grip used, but this was not studied in the present experiment.

A general conclusion is that the time when round bales are handled is not so important. Therefore, it is recommended that bales be transported to the storage area as soon as possible and ideally within the first day. With respect to the sensitivity of bales to handling, transport could wait longer, but in order to protect the bales from birds and rodents it is not recommended to leave them in the field longer than necessary.

## Acknowledgement

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# Influence of wilting and ensiling ryegrass and clover with different additives on lipid metabolism

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## Abstract

The objective of this experiment was to examine the difference in lipid metabolism in silages of ryegrass and clover, and the effect of silage dry matter (DM) content and additives. Forages were wilted to 300, 400 and 500 g DM kg<sup>-1</sup> and ensiled without additives. Forages at 300 g DM kg<sup>-1</sup> were also ensiled with a commercial inoculum and formic acid. The effect of additives on lipid metabolism was small. Formic acid increased lipolysis slightly in ryegrass silages, where it caused a decrease in lipolysis with white clover and only a trend to a decrease in silages of red clover. An effect of DM was apparent only in ryegrass, higher DM resulting in lower lipolysis. At 300 g DM kg<sup>-1</sup> no differences in lipolysis between forages were apparent, and at 400 and 500 g DM kg<sup>-1</sup> lipolysis was lower for ryegrass.

## Introduction

Most fresh forages fed to ruminants are relatively rich in omega-3 fatty acids (FA), linolenic acid in particular. Nonetheless, only a very small proportion of this linolenic acid appears in the milk or meat of ruminants. When forages are conserved as silage, the proportion of polyunsaturated fatty acids (PUFA) in meat and milk are even lower. Since PUFA are more and more being shown to be beneficial for human health, feeding strategies are investigated to reduce or even eliminate the decrease in milk PUFA due to silage feeding, e.g. by feeding linseed. However, part of the reduction can already be dealt with by adjusting the forage fed. It has been shown that feeding red clover silages instead of ryegrass silages results in meat with a higher PUFA content (Lee *et al.*, 2003). Since there were no marked differences in PUFA input between red clover and ryegrass, the difference in milk PUFA is most probably due to a difference in lipid metabolism in the silage and/or in the rumen. The goal of this experiment was to study the lipid metabolism, mainly lipolysis as it is a prerequisite of rumen biohydrogenation, during the ensiling process of ryegrass, red clover and white clover, and the possible effect of DM at ensiling and silage additives.

## Materials and methods

Red clover (Merviot), white clover (mixture of Merwi, Aberherald and Rivendel) and perennial ryegrass (Merlov) were harvested on different dates during the autumn of 2006 (18/10/06, 8/11/06 and 23/10/06 respectively). Red clover had a regrowth of 44 days, ryegrass of 40 days and white clover of 61 days. Forages were harvested using a Haldrup harvester. After mowing, the forages were wilted in a greenhouse, spread on a table and turned twice a day for uniform drying. They were wilted to a dry matter (DM) content of approximately 300, 400 and 500 g DM kg<sup>-1</sup>, which took 3, 4 and 5 days, respectively for white clover, and 2, 4 and 5 days, respectively for ryegrass. Red clover only reached a DM of 250 g DM kg<sup>-1</sup> after 5 days of wilting. To prevent excessive mould formation it was dried further in an oven at 35 °C; it took about 8h to dry to a DM of 500 g DM kg<sup>-1</sup>. Forages were vacuum packed in polyethylene bags to simulate silage fermentation on a laboratory scale. Forages were ensiled without additives, and forages at 300 g DM kg<sup>-1</sup> were also ensiled with formic acid or an

inoculum of *Lactobacillus* (Agritron BVBA, Belgium). Samples were taken after wilting and 8 weeks of ensiling for determination of the FA profile (Raes *et al.*, 2001) and the distribution over the different fractions (free fatty acids (FFA), triacylglycerides (TG), and polar lipids (PL)) (Demeyer *et al.*, 1978). Effect of DM, forage and additive were tested using ANOVA, multiple comparisons were done by the Tukey test.

## Results and discussion

**Fatty acid composition.** Herbage total FA content and content of the major FA after wilting to 300, 400 and 500 g DM kg<sup>-1</sup> are shown in Table 1. In ryegrass, as well as in white clover, there was a gradual decrease in total FA content with wilting from 300 to 500 g DM kg<sup>-1</sup>. This decrease in FA is mainly due to a decrease in unsaturated FA, indicating that oxidation could be at the origin of the observed decrease. In red clover, no significant differences between different DM contents were apparent. The different effects of wilting in the different forages could be due to differences in wilting duration.

Table 1. Fatty acid composition (g 100 g<sup>-1</sup> of fatty acid methyl esters) and total fatty acid content (mg g<sup>-1</sup> DM) of ryegrass (RG), red clover (RC) and white clover (WC) after wilting to approximately 300, 400 and 500 g DM kg<sup>-1</sup>.

	RG				RC				WC			
	30	40	50	sem	30	40	50	sem	30	40	50	Sem
C16:0	14.8	16.1*	18.5*	0.563	15.0	15.3	15.3	0.141	13.3	13.7	16.3 <sup>T</sup>	0.606
C18:2	11.2	12.3	13.6*	0.354	14.4	15.9	16.9	0.480	13.0	13.1	13.3	0.207
C18:3	64.5	62.6	58.2*	0.959	59.9	58.4	59.5	0.536	64.4	61.9 <sup>T</sup>	57.7*	1.26
Total	20.0	15.3	12.8	1.08	15.6	12.0	16.4	0.745	23.3	25.6	21.1	1.08

\*Significantly ( $P < 0.05$ ) different from control at 300 g DM kg<sup>-1</sup>, or <sup>T</sup> trend for difference ( $P < 0.1$ ).

Ryegrass and white clover were dried gradually from 300 to 500 g DM kg<sup>-1</sup>, and the decrease in FA content was also more or less gradual. Red clover, on the other hand, was dried in a short period of time (8h) from 300 to 500 g DM kg<sup>-1</sup>, and in this period no further decrease in FA content was apparent. This could indicate that for the extent of oxidation, the duration of wilting is more important than the DM content reached. After wilting to 300 g DM kg<sup>-1</sup>, white clover had the highest FA and linolenic acid content and red clover the lowest. At 400 and 500 g DM kg<sup>-1</sup> no differences between red clover and ryegrass were apparent, and white clover still had the highest content of FA and linolenic acid. Ensiling with or without additives had no marked effect on the FA composition of the forages.

**Lipolysis.** After wilting the FFA and TG fraction were not very important, as the PL fraction made up 90 to 97% of the total lipids. After ensiling for 8 weeks, the distribution of the FA over the different fractions was changed totally (Table 2). In red clover there was no effect of additives on lipolysis in the silage, although there was a trend ( $P < 0.1$ ) for a lower lipolysis when formic acid was added. In white clover silages, formic acid resulted in a significantly lower lipolysis compared to the control. In ryegrass, on the other hand, there was a significantly higher lipolysis when formic acid was used as an additive.

In all three forages, the g C18:3 100 g<sup>-1</sup> of total C18:3 in polar lipids was significantly higher when formic acid was added as an additive. Nevertheless, the effect remains rather small. The fact that lipolysis remains high or even increases when the fermentation is inhibited by formic acid indicate that lipolysis in the silage is independent of fermentation and/or pH. Furthermore, at 300 g DM kg<sup>-1</sup> there was no significant effect of forage species, implying that no inhibition of lipolysis or protection of FA could be seen in these experiments. In red clover and white clover, in contrast to ryegrass, no effect of DM on lipolysis was apparent. In ryegrass, lipolysis was significantly lower when DM was increased to 400 and 500 g DM kg<sup>-1</sup>.

Table 2. Distribution of C18:3 over the different lipid fractions (triglycerides (TG), polar lipids (PL) and free fatty acids (FFA); g C18:3 in fraction 100 g<sup>-1</sup> total C18:3) and lipolysis (%) after ensiling of ryegrass, red clover and white clover at 300, 400 and 500 g DM kg<sup>-1</sup>, at 300 g DM kg<sup>-1</sup> additives were added (no additive (C), formic acid (F) and *Lactobacillus* inoculum (I)).

			TG	PL	FFA	Lipolysis
Ryegrass	300	C	30.4	12.3	57.3	57
	300	F	6.66*	22.1*	71.3*	71.1*
	300	I	24.4	18	57.6	57.4
	400		40.3*	25.9*	33.8*	32.1*
	500		45.5*	25.4*	29.1*	28.3*
	sem (x10 <sup>-3</sup> )		9.97	9.29	8.33	11.9
Red clover	300	C	18.3	23.5	58.1	56.6
	300	F	10.9*	35.1*	53.9	52.3 <sup>T</sup>
	300	I	15.9	24.2	59.9	58.4
	400		18.6	20	61.4	59.3
	500		17	22.1	60.8	59.1
	sem (x10 <sup>-3</sup> )		2.33	4.67	13.5	3.22
White clover	300	C	16.8	23.6	59.6	58.9
	300	F	11.8*	39.8*	48.4*	47.5*
	300	I	20.1	25	55	54.2
	400		23.8	19	57.2	56.5
	500		22.9	16	61	60
	sem (x10 <sup>-3</sup> )		3.41	10.9	4.82	3.55

(Lipolysis is calculated as (g C18:3 100 g<sup>-1</sup> as FFA after ensiling – g C18:3 100 g<sup>-1</sup> C18:3 as FFA after wilting)/(100 – g C18:3 100 g<sup>-1</sup> C18:3 as FFA after wilting); \*Significantly different ( $P < 0.05$ ) from control silage at 300 g DM kg<sup>-1</sup> or <sup>T</sup> a trend to be different ( $P < 0.1$ ).

## Conclusions

In ryegrass and white clover, oxidation is probably the main factor that alters the FA composition during wilting. In red clover, no decrease in FA content could be measured, most probably due to the short wilting period between 300 and 500 g DM kg<sup>-1</sup>. No major effect of forage type and silage additive on lipolysis and FA composition could be found. These results gave no evidence of differences in silage FA metabolism between ryegrass and clover, which could be related to a reduced rumen biohydrogenation of clover compared to ryegrass PUFA.

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# Effects of multiple strains in an inoculant on the fermentation quality and nutritive value of grass-legume silage

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## Abstract

A farm-scale study was conducted to evaluate the effectiveness of multiple strains of lactic acid bacteria in an inoculant (manufactured by Medipharm AB, Sweden) on the fermentation, aerobic stability and nutritive value of grass-legume silage. The grass-legume sward was harvested at an early bloom stage of red clover, wilted to a DM content of 348 g kg<sup>-1</sup> and ensiled in round bales with either an inoculant or no additive. Inoculated and untreated silages were offered to lactating dairy cows in a feeding experiment.

Addition of the multi-strain inoculant improved the fermentation profile by lowering pH, butyric acid and ammonia-N and by increasing lactic acid ( $P < 0.05$ ). Residual water-soluble carbohydrate concentration was higher ( $P < 0.05$ ) in untreated silages than those treated with the inoculant. Improved fermentation of the inoculated silages significantly decreased ( $P < 0.01$ ) fermentation dry-matter losses. Aerobic stability was unaffected by the treatment. All components of the dairy feeding experiment, such as feed intake, milk yield and milk composition, were not significantly different between the treatments; however, for the inoculated silages these components were numerically higher.

Keywords: grass-legume silage, inoculant, dairy cow, milk

## Introduction

Natural bacteria in untreated silage ferment sugars relatively slowly and inefficiently, generating high temperatures, high dry matter losses and loss of nutrients. Obviously, selection of microbes for inclusion in an inoculant is the principal factor that will determine the impact of the product on silage fermentation and subsequent animal performance. Inoculants for silage are used to improve preservation efficiency and to prevent aerobic deterioration. However, the cause of the improvement in animal performance following feeding with inoculated silage is unclear. The extent of fermentation of water-soluble carbohydrate (WSC) during ensilage into lactic acid and volatile fatty acids can change the end products of rumen fermentation and performance of animals (Jaakkola and Huhtanen, 1993). In the majority of trials reported in the literature, the silages treated with inoculants appeared to be more digestible than the untreated silages (McDonald *et al.*, 1991). Silages treated with such inoculants may also have a positive probiotic effect on ruminant performance (Weinberg, 2004). Gollop *et al.* (2004) concluded that many lactic acid bacteria (LAB) silage inoculants have antibacterial activity and in some cases this activity is imparted on inoculated silages. This study was aimed at determining the effectiveness of multiple strains in an inoculant in improving the fermentation and aerobic stability of grass-legume silage. The study also investigated voluntary silage intake and performance in dairy cows.

## Materials and methods

The first-cut grass-legume sward (20% *Festuca pratense*, 30% *Trifolium pratense*, 50% *Lolium perenne*) was harvested, prewilted for 8-10 h, then baled and stretch-wrapped for silage on the same dates. Silages were made without any additive (C) or treated with the



multi-strain inoculant (*Lactobacillus plantarum* Milab 393, *Pediococcus acidilactici* P6 and P11, *Eterococcus faecium* M74, and *Lactococcus lactis* SR3.54), (L). The inoculant (rate of application  $5 \times 10^5$  colony-forming units  $\text{g}^{-1}$  fresh forage) was applied during forage baling. During the ensilage, samples of grass were collected to determine its chemical composition. Five additive-free bales and five inoculated bales were weighed after wrapping and after 82 days of storage, and sampled for measuring DM losses. Aerobic stability was measured using data loggers that recorded temperature readings twice a day from thermocouple wires placed in three replicated 200-g representative silage samples incubated in open-top polystyrene boxes.

Ten multiparous dairy cows between their fourteenth and fifteenth week of lactation and with an average milk yield in each treatment group ( $n = 5$  per treatment) of 23.1 (sd = 2.70) kg of fat corrected milk (FCM) at the start of the experiment were used for this experiment that lasted for 100 days. The silages were offered *ad libitum* and cows were given 280 g of a standard concentrate  $\text{kg}^{-1}$  milk. The data were analysed by one-way ANOVA, and a mean comparison by Fisher P LSD.

## Results and discussion

The activity of the inoculant was evidenced in this experiment by a higher ( $P < 0.05$ ) concentration of total fermentation acids and significantly decreased ( $P < 0.05$ ) contents of water soluble carbohydrates in the silages. The addition of multiple strains of lactic acid bacteria resulted in increased ( $P < 0.05$ ) production of lactic acid in round bale silages (Table 1). The pH value and ammonia-N concentration tended to differ between treatments. Winters *et al.* (2001) found that inoculation with *L. plantarum* improved silage quality and reduced the extent of protein breakdown during ensilage.

Table1. Chemical composition of herbage and silages and fermentation quality of silages

Treatment	Herbage (n-5)	Silages		LSD <sub>0.05</sub>	Level of significance
		C <sup>1</sup> (n-5)	L <sup>2</sup> (n-5)		
Dry matter (DM) $\text{g kg}^{-1}$	348	337	328		0.096
In dry matter $\text{g kg}^{-1}$ :					
Crude protein	133	138	140		0.823
Water soluble carbohydrates	115	52	37	12.0	0.017
Acid detergent fibre	378	352	348		0.108
Neutral detergent fibre	556	463	423		0.422
Lactic acid		28	46	12.4	0.019
Acetic acid		12	14		0.442
Butyric acid		4.7	2.2		0.881
Ammonia N, $\text{g kg}^{-1}$ total N		46	35		0.095
pH		4.58	4.17		0.077
DM losses, $\text{g kg}^{-1}$ DM		105	86	2.9	0.0005

n – number of observations.

<sup>1</sup>C = control treatment, <sup>2</sup>L = inoculant treatment.

Due to the homolactic fermentation, the nutrient (DM) losses were decreased by 18.5% ( $P < 0.01$ ) in the inoculated silage compared with the untreated one. This decrease in dry matter loss is largely due to a shift in fermentation. When fermentation is improved by the inoculant bacteria, dry matter losses from the silo decrease by 2-3 percent age units on average (Muck, 2000). There was a marginal increase of aerobic stability for the inoculated silage between the second and fifth day after exposure to air (Figure.1).

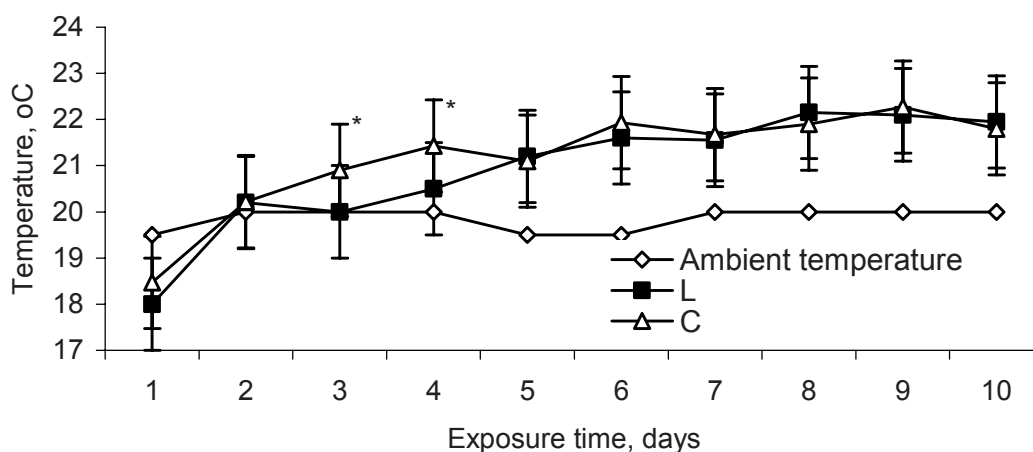


Figure 1. Changes of temperature in silages (\* denotes significant at level 0.05).

The mean silage DM intake and total daily intake with inoculant-treated silages were 12.1 (sd = 0.51) and 17.0 kg (sd = 1.47) and numerically were 0.28 kg day<sup>-1</sup> and 0.58 kg day<sup>-1</sup> higher than for the untreated silages ( $P = 0.510$  and  $P = 0.513$ ). Cows fed silage treated with multiple strains of lactic acid bacteria had a mean fat-corrected milk (FCM) yield of 19.1 kg day<sup>-1</sup> (sd = 2.24) or 1.2 kg day<sup>-1</sup> higher ( $P = 0.460$ ) than cows fed untreated silage. Treatment did not result in any significant differences in the mean milk fat and protein contents.

## Conclusions

Application of the biological additive containing multiple strains of lactic acid bacteria showed its positive effect on the fermentation process in legume-grass silage made in big bales and was efficient in reducing DM losses. Dairy cows fed the inoculated silage had a numerically but not significantly higher DM intake and FCM yield.

## Acknowledgements

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## **Session 4**

### **Can grassland-based production survive in Europe? – Pathways for future success**



# Diversity of beef farming systems and grassland use in Europe

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## Abstract

The Cattle Network Working Group of the European Association for Animal Production carried out investigations about the CAP implementation and its impact on beef farming systems. This paper develops the links between beef production and grassland in Europe from national expertise and FADN data analysis. European beef production appears highly diverse and frequently combined with other enterprises. The EU-15 territory has been divided into 7 livestock regions on the basis of bio-geographical characteristics, land use and constraints. Widely spread over the EU territory, the beef farming systems are highly implicated in the maintenance of grasslands. Thus, the multi-functional role of grasslands in providing public goods (carbon sequestration, biodiversity etc) needs to be more clearly identified. The 2003 CAP reform was a major policy change and two-thirds of Europe's beef production is now fully decoupled. Three major economic trends are changing land use: increased demand for feedstuffs, planting of bio-energy crops, and, in some countries, the total decoupling of subsidy payments. Beef enterprises are really sensitive to competition for land, particularly in areas with mixed crops and livestock.

## Introduction

It is difficult to determine how many hectares are dedicated to grassland in Europe, due to their diverse nature and the complexity for defining them as permanent or temporary pastures. From the census of 2000, 54 million ha of the 127 millions ha of the total acreage in EU 15 consist of grasslands, and 83% of these are permanent. Most of statistics show a decrease in grassland areas in Europe over the past 30 years. Quantification on 9 European countries from Eurostat data shows a loss of 18% of the grassland acreage between 1973 and 2003 (Pflimlin *et al.*, 2007). During this period, the total of the agrarian acreage has decreased leaving land to other forms of production and occupation such as forestry, industries or urban development. But it is also evident that part of the grassland area has been converted to crops. In Germany, for instance, statistics show that permanent grassland was greatly reduced (-18% in 17 years) while a part of this were replaced with cereals and oil seed crops. A halt in this decline can be observed in countries such as France since the beginning of the 2000s, where permanent grasslands remained relatively stable.

This phenomenon is not independent of the decrease in numbers of European herbivore livestock in the past and especially the rapid decline of the dairy herd since the milk-quota production implementation. EU-15 has lost one-fifth of its dairy herd in the last 10 years with the improvement of the milk cow's productivity. Part of the land left by dairy herds has been devoted to cereals and industrial crops. In the same time, the EU beef herd, which represents 40% of the bovines, remained stable. Some countries developed their suckler-cow herd in areas known for their main constraints: a non-arable soil or climate condition for instance, and it brings some guarantee for the maintenance of grassland. But the future of beef production depends on the CAP regulation. Since 2005, the implementation of premium decoupling has

left producers free to change their activity according to the market evolution. What could be their interest then in maintaining grasslands and livestock when bio-energy crops and cereals seem more profitable? Would grassland management change according to this new context?

Within the Cattle Network of the EAAP (European Association for Animal Production), a group of European economists (Beef Task force) are engaged in a common project to monitor studies for policy impact and farm strategies analysis. This group is composed of experts and researchers from the EU Member States representing some 80% of the beef production in the European Union (Ireland, Germany, Spain, Sweden, Italy and France). They gathered their national investigations in a common report divided in three topics: systems of production in Europe, CAP implementation, analysis of the first impacts on the production and land use. Beyond methodological issues (exchanges of tools, proposal for a typology of the European Beef Farming Systems), this first overview points out disparities in CAP implementation, but also the main evolutions affecting the beef farming systems: size enlargement, specialisation of farms and areas, adaptation to the market needs and eventually to the societal expectations.

In this article, we propose to come back to the link between beef production and grassland in Europe through results from the project. Starting from the description of the beef farming systems diversity and their location over the European countries and areas, we will return to their contribution to land use and the main issues they face as far as political and economic events are concerned, included the impact of the 2003 CAP implantation on the future of beef systems.

### **The diversity of constraints and land ability of the EU territory**

In the objective of defining standard procedures for monitoring and analysing economic efficiency of beef farm operations, the Beef Task Force (BTF) within the Cattle Network Working Group of the EAAP suggested a classification of the European beef farming systems. That typology allows illustrating the diversity of the farms, to localize them on the EU territory and to identify their structural evolution. The evaluation of the beef extensification payment commissioned by DG AGRI in 2007 gave the opportunity to test this farm typology on FADN 2004 data.

The classification is based on the following criteria:

- **the type of farming**, i.e. the degree of specialisation of the farm in beef enterprise, because beef production is often managed in association with other farming activities: dairy, sheep or crop production.
- **the type of cattle enterprise** – Dairy (or milk) enterprise, cow-calf (or suckler-cow) enterprise with weaning activity, backgrounding (or store cattle) enterprise, finishing (or fattening) enterprise.
- **the location** of each system.

With the FADN database, each beef farming system has been located in the European countries and placed in a zonage based on the agronomic potential of the areas. A. Pflimlin and C. Perrot proposed a division of the EU-15 territory into 7 livestock regions (See Table 1 and figure 1). These regions are distinguished on bio-geographical characteristics (boreal, Mediterranean and alpine), on land use (grasslands and part of permanent pasture, maize, crops...) and on the identification of constraints towards premium classification for mountains and less favoured areas. As herbivores use about half of the European farming territory (Vidal, 2001), the zoning described with the Eurostat Farm structure survey 2000 classified the regions according to their level of constraints and the land ability. Three large areas can be

assembled according to the beef production prevalence and its implication in the land use: zones with high levels of constraints as far as climate and agronomic potentials are concerned, i.e. Mediterranean, Mountains and Scandinavian areas, grassland areas and livestock and crop areas.

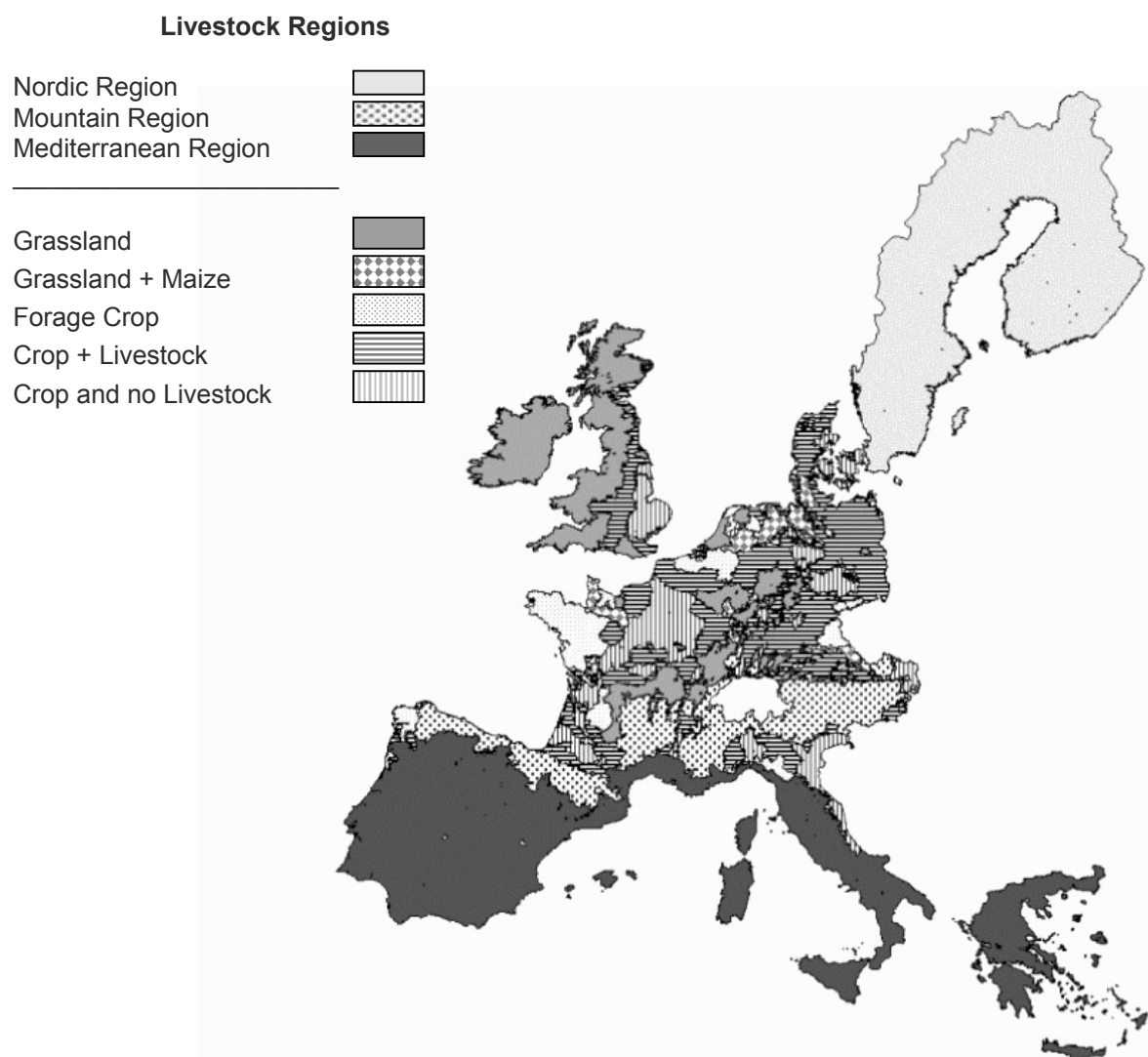


Figure 1: Main characteristics of the major livestock regions (Source: Eurostat – Structure Census 2000 adapted by the French Livestock Institut).

All in all, Mediterranean, Mountains and Scandinavian areas represent 43% of the territory and the grasslands (EU-15) and 32% of the suckler cows herds. Pastoral systems are dominant on mountain pastures with variable levels of productivity such as the wet French mountains of the Massif Central, the western centre of Spain or the hills of Toscana in Italy. This is the place for beef calving with rustic or local breeds. Herds belong to micro-farms and balance with other rural activities (tourism) in areas such as southern Italy or Greece. In the Nordic region as in mountain areas (Austria), valleys or profitable plains are dedicated to forage fodder in order to fatten some animals: bulls or heifers. Therefore, livestock play a major role in the land use and in controlling undergrowth and the risk of fire. Livestock also play a big part in sustainable development and multifunctional agriculture.

The Grassland areas include permanent pastures from Ireland, the British Isles and grassland plains of Benelux, Germany and the northern part of the French Massif Central where the climate is relatively humid. Beef activity is prevalent in those areas where grasslands and meadows represent, on average, 80% of the land use. On roughly 19% of the EU-15 territory, we find 29% of the stockbreeders and 37% of the suckler cows. Beef products are based on grass with cow-calf enterprises (France), cow-calf and steer fattening on grazing feeding on the permanent pastures (Ireland and UK). These areas remain not very intensive, as the relatively poor agronomic quality excludes higher intensification, and beef activity is often combined with sheep flocks in order to improve productivity.

The last third of the European beef herd is located in regions where agronomic soil quality allows crop production (lowlands). That means the Forage crop and grasslands and crops and livestock areas. With the oceanic climate influence, herbivore feeding is based on grass and maize silage in Galicia in Spain or in the western part of France and Belgium. It allows dairy production as well as calving and fattening of beef. The farms stand at a rather modest size, as they are relatively intensive. They often combine dairy activity with beef production and especially bull fattening. And as other livestock production is also developed (pig or poultry), they face pollution consequences and are required to respect the 170-N of the Nitrate Directive. In the plains borders and crops areas, livestock production is less present and is often managed beside field crops according to feed availability. In the Pô plain in Italy, irrigated maize cultivation is the basis of bull diet in integrated fattening enterprises. In the Sachsen plains in Germany or at the border of French plains, fattening is set down on industrial-crop by-products or some cattle herds use obligatory pastures. But here too, pollution consequences appear to be prevalent for big fattening units and their future depends highly on competitiveness compared with other production, including bio-energy crops.

Table 1. Description of livestock zones.

Major regions	UAA (ha)	Total Grassland (ha)	% Total Grassland / UAA	Perm. Grassland (ha)	% Perm. Grassland / UAA	Total Livestock Farms	% With beef cows
Total (000 ha)	126 664	54 050	43%	44 862	35%	2 319	672
Nordic	4%	3%	32%	1%	10%	3%	3%
Mountains	8%	16%	85%	17%	75%	14%	18%
Mediterranean	31%	24%	33%	26%	30%	32%	16%
Grassland	16%	31%	83%	33%	72%	16%	28%
Grassland + maize	3%	4%	57%	5%	52%	5%	4%
Forage crop	5%	6%	51%	4%	25%	9%	11%
Crop + livestock	20%	14%	30%	13%	23%	16%	17%
Crop	12%	3%	11%	2%	7%	5%	4%

Source (Eurostat-Structure census 2000 adapted by French Livestock Institute).

Table 2. Location of the EU BFS per livestock zones.

BFS (Beef Farming Systems)	Cow calf			Finishing			Dairy		Small farms	Total
	CC + sheep	CC + Fin.	Pure CC	Fin. + sheep	Spec. Finish.	small fin.	Dairy + Beef	Pure dairy		
EU livestock zones										
Livestock and crops (1)	-	3%	6%	-	1%	3%	6%	16%	3%	37%
Grasslands (2)	3%	3%	5%	1%	-	2%	4%	9%	1%	29%
Mediterranean, Mountains and Nordic	2%	1%	7%	-	-	2%	3%	12%	7%	33%
Total	5%	7%	19%	1%	1%	6%	13%	37%	10%	100%

Source EU-FADN 2004 – DG AGRI G3 adapted by French Livestock Institute (< 1% indicated by -).

(1) Forage crop, Crop + livestock and Crop areas, (2) Grassland, Grassland + maize areas.



Thus, as shown in Table 2, beef farming systems are widespread across the European territory, and this is mainly due to the ability of this production to be adapted to its climate and agronomic conditions. It is the basis of such agreement found between farms and regions in terms of suckling, weaning or fattening and specific feeding with more or less grass.

### **The diversity of beef farming systems**

Of 491,000 beef producers estimated as professional farms by the 2004 FADN data in the EU-15. 58% are cow-calf farms, 17% are devoted to fattening without rearing, and 25% are mixed dairy and beef producers. The classification illustrates the diversity of the systems and their implications for land use and production (Table 3). What is prevalent is the great combination of the beef production with other enterprises such as dairy, sheep or crops. Actually, only half of the beef producers are specialised in this activity.

The FADN-2004 counts 286,400 cow-calf producers in the EU-15. They hold the major part of the suckler cowherd and contribute significantly in the production providing weaners and backgrounders. Their contribution is effective in the EU beef production as well as in land use: they utilize 41% of the land used by the total bovine herd and with a dominance of grass and they are engaged in 41% of the production. These farms are mainly specialised in beef production with 69% of their acreage in grass, and they manage their herd at a low stocking rate: 72% retained less than 1.4 livestock unit per ha.

The cow calf producers from less favourable zones (28% of them) are located in mountain areas (wet French mountains of Massif Central, Piedmont and valley of the Alps and Pyrenees) and Mediterranean areas in the south of EU (Spain, Italy and Greece). There, suckler cows are managed in pastoral systems on rangelands and are dedicated to produce lean animals, especially weaners. Rustic breeds are mainly preferred with regard to their adaptation to the climate and the grass productivity: Aubrac, Salers in France, Piemontese or Chianina in Italy for instance. Industrial crossbreeding with meat type breeds (Charolais, Limousin) is implemented in order to improve the quality of products and the growth rate. Due to feed availability, most products (calves and store animals) are sold before long-housing periods to fatteners located in more favourable regions. To maintain their income, these farms enlarge the acreage constantly by 2% to 3% per year and increase the size of their herds. The high level of retirement and the relative low land prices facilitate that permanent extension and the reduction of the stocking rate.

Cow-calf and backgrounders (store cattle) producers are dominant in the grassland areas of France, Belgium, Irish and British Islands and Sweden. On those areas, with regular rainfall, grazing systems throughout the year with little supplementation can cover feeding needs. The agronomic context limits cultivation ability and especially wetness or slope. Therefore, feeding systems are based on permanent grassland with moderate intensification. In the French grassland plains, larger herds are retained with Limousin or Charolais breeds and weaners are dedicated to fatteners from Italian or Spanish units. In Ireland and Scotland, cow-calf production is often combined with sheep production and backgrounders are sold for local fattening. Most stockbreeders are part-time farmers. Thus, the maintenance of grassland areas is justified to maintain livestock farming activity on personal land with little investment.

Table 3. EU Beef Farming System (BFS) characteristics.

Production BFS (Beef Farming Systems)	Cow calf (CC)			Finishing (F.H)			Dairy		Small farms	Total
	CC + sheep	CC + Fin.	Pure CC	Fin. + sheep	Spec. Finish.	small fin.	Dairy + Beef	Pure dairy		
FADN farms	1305	1358	3182	194	350	1016	3420	9706	1077	21 608
Extrapolation (1000 farms)	49.9	65.1	171.4	9.2	12.7	58.8	123.8	338.7	91.4	920.9
Acreage (ha)	151.9	73.3	69.7	87.9	94.0	45.9	81.5	56.9	19.8	63.10
% grass on total acreage	88%	63%	69%	68%	45%	42%	62%	61%	33%	62%
% beef on total returns	34%	49%	53%	22%	51%	34%	21%	9%	14%	19%
Labour unit	1.7	1.5	1.4	1.6	2.0	1.4	2.0	1.9	1.4	1.7
Livestock unit	52.5	61.4	47.4	39.5	127.2	31.6	93.6	66.7	3.8	58.1
% BFS farms	5%	7%	19%	1%	1%	6%	14%	37%	10%	100%
% land (on land for BFS)	13%	8%	20%	1%	2%	4%	17%	33%	2%	100%
% BFS livestock unit	5%	8%	15%	1%	3%	3%	22%	42%	1%	100%
% Beef Production (1)	6%	11%	24%	1%	6%	7%	20%	24%	1%	100%
Stocking rate (LU ha <sup>-1</sup> )										
< 1.4 LU ha <sup>-1</sup>	65%	50%	72%	29%	15%	56%	31%	34%	60%	47%
> 1.8 LU ha <sup>-1</sup>	12%	23%	13%	23%	57%	25%	34%	37%	27%	28%
%OTEX 41-42-43-44-71	85%	63%	66%	57%	53%	45%	84%	86%	25%	71%

Source EU-FADN2004 – DG AGRI G3- adapted by French Livestock Institute.

(1)In value: the contribution of the different types has been calculated in regard of the acreage used (total and grass), the total of livestock unit and the total of beef outputs (in value (€) and net of purchasing).

Cow-calf and young bull producers from livestock and crop zones are situated in the west of France, Limousin region and French or German plains borders. The bovine herd is kept on farms engaged in fattening the progeny as suckled calves, bulls and heifers. This fattening enterprise is designed to add value to the weaning activity. With a rather bigger size, they manage land more intensively with production of forage crops, and 23% operate at a stocking rate greater than 1.8 LU ha<sup>-1</sup>. Here, total grasslands always represent 63% of the acreage, but temporary pastures prevail and are combined with maize silage. Implicated in the providing of local beef industries, those systems face competition for land with other livestock activities and crops. In some regions (Brittany, Flanders) the implementation of the Nitrate Directive limits also extension of the herd size and restricts farmers in intensification.

Cow-calf and steer producers from the Irish and British islands combine beef and sheep production. In Ireland and Scotland, where permanent grassland and wet rough grazing cover most of the territory, cow-calf production is often combined with sheep production and stock are sold or fattened as 24-month-old steers or heifers according to the market price. This management on grass feeding is helped by the calving concentration in springtime but needs some housed-fattening with grass silage and cereals. Independently of the relatively large area of the farms (151 ha on average), most of those breeders are part-time farmers and have two jobs. So, livestock activity first depends on cost of land and labour availability.

The FADN census does not give a good overview of the fatteners' enterprise. Relatively few in number, the farms specialised in finishing activity are involved in 14% of the EU beef production. They are the result of a progressive localisation of production: calving in less favourable zones, fattening where crops and maize silage are available. Among them are 12,700 farms that are specialised in bull fattening. They are involved in 36% of the livestock units and play a significant role in the beef production of Italy, Germany, UK and Spain.

Located in favourable areas for crops, those systems are facing different issues: reduction in environmental risks, increase in feeding prices and competition with bio-energy crops.

Finally, 123,800 enterprises are involved simultaneously in dairy and beef production. Those systems represent one-quarter of beef producers and use 17% of the beef farming land. In most cases, the beef production has been developed on farms with small milk quotas. Different types of production occur according to land and labour availabilities. It may be bull fattening of dairy calves on the French or German farms, or it may be steer and heifer production on pastures in Great Britain and Ireland. Their weight in the EU beef production is determinant (20%). With the liberation of 2% of milk quota and in the new context of decoupled payment, they can choose to specialise their farm on dairy enterprise in order to simplify the livestock management.

### **The implication of the beef farming systems in the grassland maintenance**

According to FADN data, the beef farming systems use about the half of European grasslands. 62% of their own farm acreage is dedicated to grasslands. It's quite similar to the needs of dairy systems pasture acreage but this employment is shared between beef breeders along the chain of production: from the calf rearing mainly managed on pastures and beef fattening often based on grain or maize silage diet. This segmentation is in accordance with the agronomic and climate context (calving period, feedstuff production or purchase, ability in fattening, etc.), and explains localisation of calving in some areas and concentration of fattening in others. The case of the Irish and British beef farming systems need to be pointed out as real grassland based beef production with rearing and finishing of steers and heifers 'on the farm' and with a self sufficient feeding on pastures. This management is specific to favourable grasslands areas.

Because forage system is a major part of the global farming system, the use of grasslands interferes in the profitability of beef production. According to our monitoring of beef cost-of-production (French Livestock Institute, 2007), feeding costs represent between a third to the half of the total beef costs but the part left to grass in the diet remains a determinant in the beef farming sustainability. The international beef cost-of-production comparison available from the agri benchmark Beef & Cow-Calf Network (Federal Research Institute for Rural Areas, Forestry and Fisheries, Johann Heinrich von Thünen Institute, 2007), points out the gap of feeding cost from 1 to 3 between South American and European beef enterprises. In Brazilian systems for instance, total grazing fed accounts for a decrease in the feeding cost by 20 € per 100 kg of meat produced, whilst it rises to 70 € per 100 kg in some European beef systems. Furthermore, the pastoral management implicates a lower demand of other inputs and equipment (housing, machinery etc.). These do not explain all the differences of competitiveness. Other components of the concurrency are labour, land and capital costs, factors that lead the choice of livestock management. This last part is the Achilles' heel of EU beef costs when farms remain rather small in size and still organised around family labour and capital.

### **The beef production involved in the multi-functionality of grasslands**

After different livestock health crises in which livestock, and more precisely beef production have been involved, environmental and societal issues of the land quality, water and air quality become stronger concerns nowadays. At this point, the role played by meadows in the production of public goods appears to be significant. Teagasc (Dune, 2004), showed that public goods like food safety, animal welfare, and environmental and ethical issues need to be explained and taken in account as society becomes more aware of the external costs involved

in food production and maybe could afford to place a higher value on it. The French Livestock Institute (Pflimlin and Perrot, 2007) defines those public goods as goods available for everybody and without special quantitative or qualitative limit. Those public goods escape the logic of the market; it is necessary to develop another type of strategy to develop them, and for that reason the part play by meadows needs to be assured by public policies and financial assistance.

Various works for the cartographic study of the French Livestock Institute (2000) confirm the positive effect of meadows and forest on the quality of water. The lowest nitrate concentrations in surface water are observed in the areas where permanent meadows cover more than 70% of the SAU. The stocking rate is moderate in these areas (around 1 LU ha<sup>-1</sup>) and the mineral nitrogen fertilisation is modest (less than 50 kg ha<sup>-1</sup>). The nitrate content in water is often below 10 mg litre<sup>-1</sup>. The meadows also have an effect on reducing the contamination of water by residues of plant protection products. It's due to the few need of adventitious control on permanent pastures, and reduction of infestation on farming rotations that are integrated with meadows. Finally, the meadow cover reduces the surface run-off and with it the movement of compounds towards the rivers. This explains the value of the establishment of grass areas along rivers which is required within the framework of the Nitrates Directives and the cross-compliance of the CAP.

The meadow stores carbon. A recent synthesis by the French INRA (2002) confirmed by many other bibliographical references, shows that on the first 30 cm of ground, the quantity of carbon is of 70 tons per hectare for the permanent meadows, against 45 tons per ha for the grounds in annual cultures, with strong variations. These differences are explained by a more important carbon flow in the meadow systems (more roots and plant remains), a slower decomposition of the organic matter in the absence of ploughing and aeration of the ground, and finally, a weaker degradation of the meadow roots, richer in lignins and polyphenols. Thus, any change in the land use implicates a significant rise or destocking of carbon. Article 3.4 of the Kyoto protocol admits the storage of carbon under the meadows in order to control greenhouse gas emissions, but this is not yet considered at the European level. According to evaluations of INRA, the storage of carbon compensates for half of the ruminant emissions related to the livestock transforming meat and milk grass: methane, nitrogen oxide and nitrogen. Other sources of storage need to be taken into account such as the contribution of manure as well as hedges, ditches and woods in terms of their organic matter storage.

Facing new societal issues, the role of grasslands appears significant in the maintenance of biodiversity and landscape management. Most of the meadows have a good vegetation diversity compared with mono-cropping. This is due to the long-term pastoral practices, which guarantee the maintenance of a large variety of grass species but also insects, microorganisms, and other fauna such as earthworms. Here too, the implementation of new Habitats and Birds Directives within the framework of the CAP symbolises the emergence of this matter. The experts also insist on the connection between meadows and hedges, and its positive impact on biodiversity. Furthermore, in the Mediterranean basin, pastoral breeding is known to keep landscape open and limit undergrowth. In this way, it plays an essential role in reducing the risk of fire and avalanches in mountains.

After several food health crises and more precisely the BSE crisis related to intensive feeding practices, grass feeding had been presented as a condition to reinforce the guarantee given to the consumer about food safety. Since then, the positive image of the meadow is a part of the sales points in the stores and it is included in the management required for labelling or organic

production. It is true that research results prove the positive role of grass feeding to improve nutritional and sensory qualities. Rich in omega-3, the grazed grass improves the content of the meats of unsaturated fatty acids and it contributes to intensify its flavour.

These environmental and qualitative assets of the meadow are summarised in Table 4 and explained in the Annex 2. They illustrate the multi-functionality of the grassland systems. A gradient exists, which goes from annual fodder and the meadow on an intensive mode to the extensive permanent and wet meadows. The production of public goods appears to be more important as far as extensive grassland management is engaged: in terms of organoleptic qualities of the foodstuffs, and in the fight against erosion or the regularisation of water quality for instance. But actually, those by-products are too far removed to find an appreciation on the beef market according to the high prices of meat in Europe and the relative scarcity of protected designation of origin (PDO) in beef.

Table 4. The multi-functionality of grasslands.

Type of grass	Practices	Biodiversity	Landscape affect	Water quality	Erosion prevention	Carbon storage	Products quality
Annual fodder	(1)	*	*	*	*	*	*
	(2)	*	*	**	*/**	*	*
Temporary meadows	(3)	*	**	*/**	**	**	**
	(4)	*/**	**	***	***	**	***
> 5 years or permanent meadow	(3)	**	***	**	**	***	**
	(4)	***	***	***	***	***	***
Grasslands with ecologic sensibility	(5)	*/***	****	*****	***	*****	*****

Source: French Livestock Institute-2007.

(1)maize with no crops alternating (2)maize with crops alternating (3)intensive management (4)reasoned fertilisation (5) wet or dry meadows.

\*little impact, \*\*\*\*\*high impact.

### CAP 2003: more place for subsidiarity

For some years, environmental considerations have been one of the main aspects of the CAP. In its first steps, the CAP was based on the desire to guarantee self-sufficiency in basic foodstuffs and after the 1990s a new orientation have been engaged with the policy of subsidiarity that links the production with respect of environmental, food safety and animal welfare standards. The second pillar, a special fund engaged to support those standards has been promoted with the extension of a global subsidies modulation. After the main diseases crisis (1996-2001), and in order to recover consumer confidence, this orientation was reinforced with the so-called Mid-Term Review (Luxembourg, 2003). This gives more place again to subsidiarity and proposes the decoupling between premiums and production.

Actually, the CAP-reform of 2003 is a major change in agricultural policy. Main characteristics are the decoupling of the direct payments from actual production (beef, suckler-cows, cereals, milk, etc.) and the linkage of the payments to the fulfilment of regulations regarding the maintenance and management of the land and environment (cross compliance). If a recipient of payments does not comply with the regulations, payments may be cut or even withdrawn.



These general principles have been modified in many countries of the EU. The result is a co-existence of different ways of implementation. Some countries have opted for keeping some direct payments partially or fully coupled to the animals maintained or produced. Some have chosen the full decoupling of payments with the 'single farm payment' (SFP) based on the farm-individual historic payments or an acreage-based payment.

Finally, two-thirds of the beef production in Europe appears to be fully decoupled. It can be assumed that at least for male animals the low levels of payments remaining coupled will not lead to different decisions about continuation or stopping production when compared with the fully decoupled situation. However, the situation of the cow-calf production is somewhat different. Payments for two-thirds of the suckler-cows at the EU-level remain fully coupled, mainly because the two dominant suckler-cow countries in Europe, France and Spain, have opted for that system. The suckler-cow premium is €180-200 per cow, a level that will most likely have a significant impact on the decision whether to continue cow-calf production or not.

### **Specific measures to maintain grasslands**

As far as Member States could have chosen different implementation of the 'Mid-Term Review', the CAP looks like an 'à la carte' policy. Except for the specific 'policy for grass' as applied in France and certain regions, grasslands are not a specific target for the CAP but will be affected by evolution of beef premiums and cross compliance.

The acreage-based payment was proposed because it simplifies the premium sharing between farms. It also appears as rebalancing between cultivated fodder and grass. In the dynamic 'hybrid decoupling model' adopted in Germany, the grasslands payment will gradually increase after 2009 to reach the same level as the croplands payment while the single farm payments will be phased out. By favouring grassland, the choice could weaken the activity of beef finishing in intensive livestock farms, but the delay to full implementation should allow these farms to adjust the management and the size to the new regime.

Related to the cross-compliance obligations, the 'Maintenance of permanent pasture' directive limits the ability to convert permanent grasslands (more than 5 years old) to cropland. Actually, this measure affects farmer's practices only in the case where the national or regional share of permanent pasture is decreasing significantly (more than 5% compare to reference years).

The implementation of the article 69, gives also the opportunity to State Members to orientate some premiums to grasslands and beef production. Since 2003, they can take up to 10% of the envelope of the Single Farm Payments to use it as an 'additional payment' with 'particular types of agriculture which are important for the protection of the environment or for the improvement of the quality and the marketing of the agricultural produce'. In Spain for instance, that article gives the opportunity to support the extensive beef holdings and maintenance of livestock activities involving a benefit from an environment point of view. It is oriented to suckler cow holdings and the farms that have a maximum of 1.5 LU ha<sup>-1</sup>. In 2006, 85% of Spanish suckler cows have received this additional payment. Same types of measures have also been deployed in Scotland or in Italy.

Through the Pillar II, measures for pastures remain limited by financial restraints and evolution of the budget. Since 1992, some countries have proposed incentive Agri-Environmental Measures for the maintenance of permanent grasslands and good farming practices. This is

the case of the 'premium for grass' in France that concerned about 5 million ha and two-thirds of French beef breeders, in spite of a quite modest premium per hectare and reconsideration of management practices. Other countries or regions such as Austria, Bavaria and Baden-Württemberg in Germany, and Sweden have proposed similar premiums but these ambitious regional policies necessitate considerable means. The Irish Rural Environment Protection Scheme and British Environmental Stewardship also lead farmers to respect good agricultural practices: to keep their meadows and limit the fertilisation.

### **Grasslands use impacted by new economic trends**

According to the analysis of the Beef Task Force of EAAP, it seems that the beef farming sector will go on with the structural evolution of specialisation and enlargement related to the disappearance of smaller farms, less specialised and less competitive. This phenomenon occurs in certain countries as France, Spain or Sweden due to the higher rate of farmers retiring. It means that land management is a major issue for the future, as well in crops areas than in grasslands areas.

Moreover, three main economic trends will probably change the land use: first, the greatly increasing demand for foodstuffs in cereals, dairy and beef products, and more or less related, rising inputs costs; secondly, the emergence of the production of bio-energy crops that compete with food crops and the needs of herbivores for land, and finally the implementation of total decoupling in some countries. Those events will impact differently on different areas and consequently on the beef farming systems. It will depend both on the potential alternative uses and on their profitability.

In the 'Less favourable areas' of France and on the central plateau in Spain, generally disadvantaged or mountainous, there is no alternative than beef except forestry or sheep production. The decision to maintain the suckler premiums coupled payments continues to support the extensive cow-calf production and the maintenance of rangelands. In those areas, fattening heifers or young bulls and producing steers would be less profitable than before. Moreover in Spain, the implementation of article 69 about the support of extensive holdings ( $< 1.5 \text{ LU ha}^{-1}$ ) should help to maintain a livestock activity in those regions. Those farmers face also the recent decreasing of the weaner prices. This context is due to the high growth of input cost for fattening enterprises (from Spain or Italy) and market stress related to the Bluetongue disease. With relatively low costs of production, those beef farming systems should improve again their pastoral management in an enlargement context. However, their sustainability will remain dependant from 'Less favourable areas' allowances and Agri-Environmental Measures.

In grassland areas, the land availability is the main matter. When ploughing is not attractive, grass feeding appears as an advantage for competitiveness of beef production in a period of increasing feeding costs. In the French grassland plains, it can help the farmers facing the demand of the importers to adapt their production: type of weaners, season of production, calving period, and guarantees of production. In the Irish and British islands, the suckler cowherd was not too much affected by the total decoupling until a small recent decline. However, marginal enterprises led by part-time farmers seem sensitive to other alternatives in land use such as city implantation. Teagasc underlines the impact of the cereal price increasing on their farms. Reverse to the increasing use of grain in Irish beef fattening in recent years, this will lead beef fatteners to develop production systems more based on grass. This will have two negative consequences, namely increase methane emissions and increased



seasonality of production. The Nitrates directive and associated environmental regulations could also limit the development of large-scale winter fattening units in Ireland.

The implementation of a combined model of CAP in Germany needs to be pointed out. The introduction of the grassland premium and the increase of this premium to the same level as cropland premiums (around €300 per ha) in 2013 is an indirect – and nevertheless decoupled – support for grassland systems. The decision to continue suckler-cow production firstly depends on the alternatives available to fulfil the cross-compliance requirements. Mulching the land is the new reference system for all agricultural activities. But if livestock production is costing more per ha than mulching, it is not the profitable alternative. Moreover, the introduction of the grassland payments will lead to an increase in rental prices, thereby diminishing whole-farm profitability, especially in Eastern Germany, where the rented land is dominant.

And finally, beef enterprises seemed to be highly sensitive to the competition for land in livestock and crop areas. Some 37% of the beef farms and 32% of the EU suckler cow herds are located in those regions where a third of the grassland acreage is cultivated. Most of those arable grasslands are not protected by the "Maintenance of permanent pasture" obligation and could easily be converted to crops. In 2005 and 2006, the balance between the sale price of bulls and the purchase price of the weaners was in favour of fattening, compared to crop production, but the balance has changed in 2007 and is reinforced with the increase of inputs costs. Crop prices have now risen in response to the recent increase in demand for energy crops, firstly in America but also in Europe. The Federal Research Institute for Rural Areas, Forestry and Fisheries, (Johann Heinrich von Thünen Institute, 2007) showed that intensive bull finishers from Sachsen plain are those who will face significant income losses with the single farm payment implementation. Moreover, the recent expansion of bio-energy plants will reduce the profitability of beef production as it will contribute to the increasing of land prices and the rising of the opportunity costs for feeding corn silage. In those areas, most of the beef farms are combined systems with cow-calf and fattening or dairy and fattening production. Definitely, the future of those beef activities will depend on their capacity to improve again competitiveness as far as calves production and feeding self-efficiency are concerned.

## Conclusion

In the proposal for the 'Health Check' of the CAP reform, the agricultural Commissioner explains how 'the CAP increasingly contributes to heading off the risks of environmental degradation and to delivering many of the public goods that our societies expect'. Actually, this objective needs to be comforted as regards of the constant decreasing of the permanent grassland areas, which can provide such public goods. As far as the beef sector is concerned, the lack of harmonisation in the MTR implementation is obviously the symbol of the difficulties to find incentive allowance adapted to the diversity of producers spread over the EU territory, in favourable and in less favourable areas. In the current economic context, it seems that the beef production will highly be affected by the growing demand for cereals and bio-energy crops and the new competition for land. Located in crops areas, fattening enterprises are more sensitive especially in the situation of beef premium decoupling and with the ability to balance between beef production and crops. However, the production of weaners or backgrounders from grasslands areas could also be impacted by the prices recession.

European beef farmers will definitely never be competitive with Brazilian producers whose cows graze all year long with no housing or forage stores and with costs of production three

times cheaper than ours. Part of the explanation belongs to the size of the European beef farms and to the cost of labour, land and capital factors. That is why structural evolution in this sector will be particularly long to be implemented. However, this new situation is clearly determinant for the future of the European beef production. Little competitiveness can be gained with the improvement of the self-efficiency, especially in beef farming system based on grass feeding. Nevertheless, the role played by the beef production in the maintenance of half of the European grasslands needs to be more identified as well as its contribution in the providing of public goods.

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## To graze or not to graze, that's the question

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### Abstract

Current trends in livestock farming in Europe are causing a decline in the popularity of grazing systems for dairy cows. This paper presents an overview of dairy cow grazing in northwest Europe. The grazing system affects various aspects such as grassland productivity, animal welfare, environment, economy, labour and even society. Grazing has advantages and disadvantages. The importance attached to the various effects of grazing is very personal. It is shown that grazing for a limited part of the day scores well on the whole. In the end, the personal motives of the farmer will answer the question 'to graze or not to graze'.

Keywords: animal welfare, dairy cattle, grass intake, grazing, grazing system, sustainability

### Introduction

Throughout Europe, forage is the main feed for dairy cattle. Grass is fed either fresh – predominantly through grazing – or in a preserved form as silage or hay. There is a trend however towards less grazing in most European countries. The reasons for this vary but are mainly due to the different types of farm systems that typically occur in different regions.

Modern, large-scale farms with high yielding dairy cattle, such as those increasingly occurring around Europe, may reduce grazing in order to control the diet and optimise grassland utilization. Unquestionably, dry matter intake (DMI) and hence nutrient intake of dairy cattle fed grass only is limited. Van Vuuren and Van den Pol-van Dasselaar (2006) used data of Bruinenberg *et al.* (2002), Bargo *et al.* (2003), Butler *et al.* (2003), Ribeiro Filho *et al.* (2005) and Tas *et al.* (2005) to calculate that when fed a grass-only diet, a maximum DMI of 110 to 120 g (kg body weight)<sup>-0.75</sup> can be expected. This is enough to meet the requirements of maintenance and 22 to 28 kg of milk. Cows with higher milk production capacity require supplementary feeding to meet their relatively high energy and protein requirements. However, in situations where supplementation is offered grazing time decreases.

In addition, increased herd sizes make grazing more difficult. In general, when farms increase in size, the grazing area (i.e. land base) in the vicinity of the farm remains the same, which leads to an increase in grazing pressure through increased stocking rate. When additional land is incorporated into the grazing area, walking distance to the milking parlour may become a limiting factor.

Another reason for less grazing is the increasing use of automated milking systems. It is possible to combine both grazing and robotic milking systems (e.g. Wiktorsson and Spörndly, 2002), it can, however, be problematic (Parsons and Mottram, 2000). Information collected through a survey conducted in the Netherlands revealed that only half of the farms using robotic milking systems practised grazing (Van Dooren *et al.*, 2002). However, on the other farms surveyed about 90% turned cows out to pasture to graze. Over the next ten years we expect a sharp increase in the number of European farms with robotic milking systems.

In some countries, grass growth has been reduced during the summer months over the last number of years, especially in July and August. The uncertainty regarding the availability of grass supply may be another reason for less grazing. Finally, the need to reduce mineral losses

and increase labour efficiency may also be reasons to cease grazing. Especially the latter is an important factor for many farmers.

From the reasons outlined above it is clear that current trends in livestock farming in Europe are causing the decline in the popularity of grazing for dairy cows. Is this a matter of concern? Is grazing important and, if so, why? Grazing sends out a signal about dairy farming. The general public in Western European countries is increasingly calling for farm systems in which animals can display their natural behaviour, and grazing is an important aspect of such behaviour for dairy cattle. In addition to issues related to animal welfare, an open landscape with grazing cattle is highly appreciated by the general public. The use of systems that involve only restricted or no grazing opens up the opportunities for moving away from grass as the major feed, with possible large effects on production, economics, environment and landscape. In several countries large projects to stimulate grazing have already started or are being prepared at the moment, for example in Luxembourg ([www.fill.lu](http://www.fill.lu)), the Netherlands ([www.koewij.nl](http://www.koewij.nl)), Switzerland, Austria, Germany and the UK. The aim of the FILL-pasture project in Luxembourg is to promote grazing. The aim of the Koe and Wij project in the Netherlands is to make farmers aware of the different aspects of grazing and let them decide for themselves using arguments with respect to economy, labour, environment and personal preference.

To graze or not to graze, that's the question! This paper provides information with regard to answering this question. We will describe the current situation and trends in Europe. We will examine some problems relating to grazing: the dilemma of a high herbage intake versus high utilization and the increasing walking distances for dairy cows when farm size increases. We will consider the effect of different grazing systems on aspects such as sustainability, the animal and the society. Finally, the determining factors for grazing will be discussed.

## **Grazing in Europe**

There are few long-term data on grazing available in Europe. However, scientists of several countries in northwest Europe indicate that grazing is decreasing in importance. There is large variation both between and within countries. In Denmark, 16% of dairy cattle did not graze in 2001. By 2003 this number had increased to 30% and it is still increasing. The countries Norway, Sweden and Finland have welfare legislations stating that cattle must have access to pasture or alternative exercise areas outdoors for a minimum period of time during the summer (six weeks to four months depending on location). However, there are no requirements regarding the contribution of pasture to the total energy supply. In Norway, 10-25% of the yearly net energy supply comes from pasture. The grazing season in Norway and Finland varies from less than 100 days in the mountain areas and far north to approximately 180 days in the coastal areas in the south. In Finland grazing became less important from 1990 onwards until the introduction of aforementioned welfare legislation in 2006. In the Netherlands the number of grazing dairy cows has been monitored rather intensively from the early 1990s onwards (Figure 1). Especially in the last few years the number of dairy cows which are kept indoors for all or part of the summer has increased considerably. If grazing is practised, the average number of grazing hours  $\text{cow}^{-1} \text{ day}^{-1}$  has reduced. In Germany there is a similar trend. In Luxembourg, it is estimated that up to 10% of the national herd does not have access to pasture; also the number of grazing animals is decreasing. A survey conducted in 2005-2006 in the west of Belgium revealed that at least 4% of the farms dairy cattle do not graze. When asked, 13% of the farmers said that in future dairy cattle will not graze on their farms. In the UK, it was estimated that less than 5% of the dairy cattle did not graze in 2005; this number is increasing. In Ireland, grass-based seasonal systems of milk production predominate. The length of the grass-growing season varies from about 8 months in the northeast to up to 11 months in the extreme southwest.

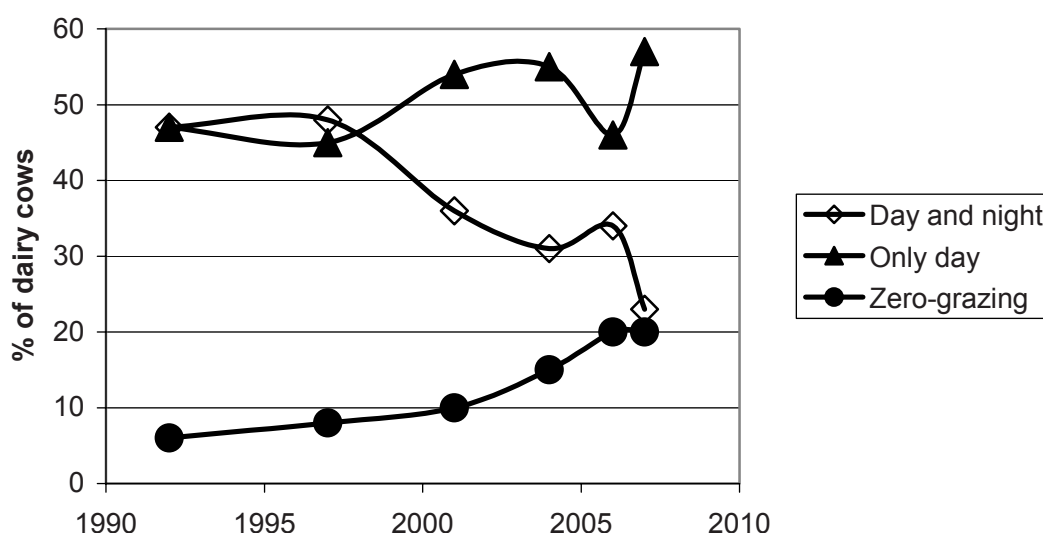


Figure 1. Grazing systems in the Netherlands in the period 1992 to 2007 (% dairy cows) (CBS StatLine databank, <http://www.cbs.nl/nl/cijfers/statline/index.htm>).

Throughout northwest Europe there are not only considerable differences in the length of the growing season, but also in dry matter yield. Compared to these large differences, the differences in nutritive value of herbage are smaller. The net energy is high in spring, with more than 7 MJ kg DM<sup>-1</sup>, this decreases during the summer period and tends to increase slightly during late summer and autumn (Table 1). The crude protein level follows mainly the same pattern as the net energy. It is high in early spring and again in late summer and autumn. The data of Table 1 are partly from extensive surveys and partly from experiments. It is therefore not possible to draw conclusions about the differences between countries.

Table 1. Net energy and crude protein content of herbage for dairy cows during the growing season in Ireland (Horan and Shalloo, 2007), the Netherlands (Van Vuuren and Van den Pol-van Dasselaar, 2006) and Norway (Johansen, unpublished data).

	Mar	Apr	May	June	July	Aug	Sept	Oct/Nov
Net energy for lactation (MJ kg DM <sup>-1</sup> )								
Ireland	7.1	7.1	7.0	6.9	6.6	6.4	6.7	6.7
The Netherlands		7.2	7.0	6.9	6.8	6.8	6.9	
Norway			8.8	7.2	6.9	6.9		
Crude protein (g kg DM <sup>-1</sup> )								
Ireland	223	222	166	176	169	189	203	228
The Netherlands		237	214	223	227	237	261	
Norway			264	207	193	227		

### The dilemma of a high herbage intake versus high utilization

The productivity of dairy cattle is influenced by herbage intake and by nutritive value of the herbage. Several management factors affect dry matter intake (DMI) (e.g. Dillon, 2006). A very effective way to increase herbage DMI is to increase herbage allowance. Data from Johansen and Höglind (2007) illustrate this. When 12 to 24 kg DM allowance was offered, DMI increased by 0.24 kg for each extra kg DM of herbage allowance. Sward utilization decreased, however, from 72% at 12 kg DM herbage allowance to 51% at 24 kg DM allowance. This decrease in herbage utilization is a matter of concern.

Although herbage intake can be increased by offering larger allowances, the negative effect of higher residuals in subsequent grazings is also clear (Taweel, 2006). Research at Moorepark



in Ireland showed that pastures grazed to a post-grazing sward surface height of 5.5 to 6.5 cm in the May to June period compared to a post-grazing sward surface height of 8.0 to 8.5 cm achieved a higher DMI ( $+ 0.8 \text{ kg day}^{-1}$ ) and higher milk production ( $+ 1.2 \text{ kg day}^{-1}$ ) in the July to September period, due to a higher proportion of green leaf and lower proportion of grass stem and dead material.

Allocating herbage in early spring may positively affect herbage intake. Kennedy *et al.* (2005) showed that dairy cows in early lactation, that were turned out to pasture full-time post calving, produced the same amount of milk as cows that remained indoors until early April, but with a lower fat content (38.6 versus 41.6 g kg<sup>-1</sup>) and higher protein content (33.6 versus 30.7 g kg<sup>-1</sup>). The cows on the early spring grazing system continued to maintain a higher milk protein content and higher grass DMI than their indoor counterparts for 12 weeks after the experimental treatments were no longer imposed. O'Donovan *et al.* (2004), in an experiment carried out in France, also showed that early spring turnout to pasture has positive effects in subsequent grazings. During the mid-April to early July period pastures initially grazed in early spring (February and March) produced swards of higher quality and higher milk production potential than swards initially grazed in mid-April. The positive effects of early grazing are due to a higher leaf proportion and hence greater digestibility compared to later grazed swards.

Kristensen *et al.* (2007) showed that restricting grazing time forces the cow to graze more efficiently, although the reduction in herbage intake and animal performance cannot be fully compensated. Comparison of animal production in a system of restricted grazing with supplementary feeding and a system of full-time housing showed that a high milk production of 9000 kg cow<sup>-1</sup> yr<sup>-1</sup> can be realized in both systems (Beeker *et al.*, 2006).

Also other strategies have been developed to realize a low post-grazing height. Mayne *et al.* (1988) used a leader follower system, with high and low yielding groups of dairy cows. Also systems with heifers and sheep as followers have been used. Furthermore, topping has been advocated (Stakelum and Dillon, 1990) to maintain sward quality mid-season. Recent experiments on peat soils did not, however, show a positive effect of topping (Holshof *et al.*, 2006).

We conclude that there are opportunities to increase the DMI of grazing dairy cows; however, stringent management factors need to be put in place. The main objective is to find the balance between a high herbage intake, little residual herbage and little negative side effects later in the growing season.

### **Walking distance**

Grazing becomes more complicated with increasing herd size. The area that is needed for grazing increases proportionally with the herd size (Figure 2). With increasing herd size, the average distance between paddock and milking parlour increases too. To calculate the average walking distance, we simply assumed a square farm area with farm buildings in the corner and with square paddocks at a depth: width ratio of 2: 1. Walking speed of dairy cows was assumed to be 4 km hr<sup>-1</sup>. This implies that it takes 15 minutes to walk 1000 m. In farmers' questionnaires in the Netherlands, 1000 m is seen as an acceptable maximum walking distance for dairy cows. Figure 2 shows that under these conditions 30 ha is the maximum area that can be grazed. A dairy herd of 150 cows can graze on 30 ha during the whole growing season with 8 kg DM of maize supplementation during the night period. With larger herds, e.g. with 250 dairy cows, grazing during the whole growing season is not possible on an area of 30 ha. A different position of the farm buildings, e.g. in the centre of the farm area, can provide much more grazing opportunities. For example a dairy herd of 600 cows can in that situation graze on 120 ha during the whole growing season with 8 kg DM of maize supplementation during the night period. However, in several countries it is not easy to realize farm buildings in the centre of the farm area.

The possibility of milking dairy cows at pasture is reconsidered again. Oudshoorn (pers. comm.) tested a system of an in-field milking robot in Denmark in 2007. Experiments with a mobile robot or an easy transportable milking parlour will probably start in the Netherlands in 2008.

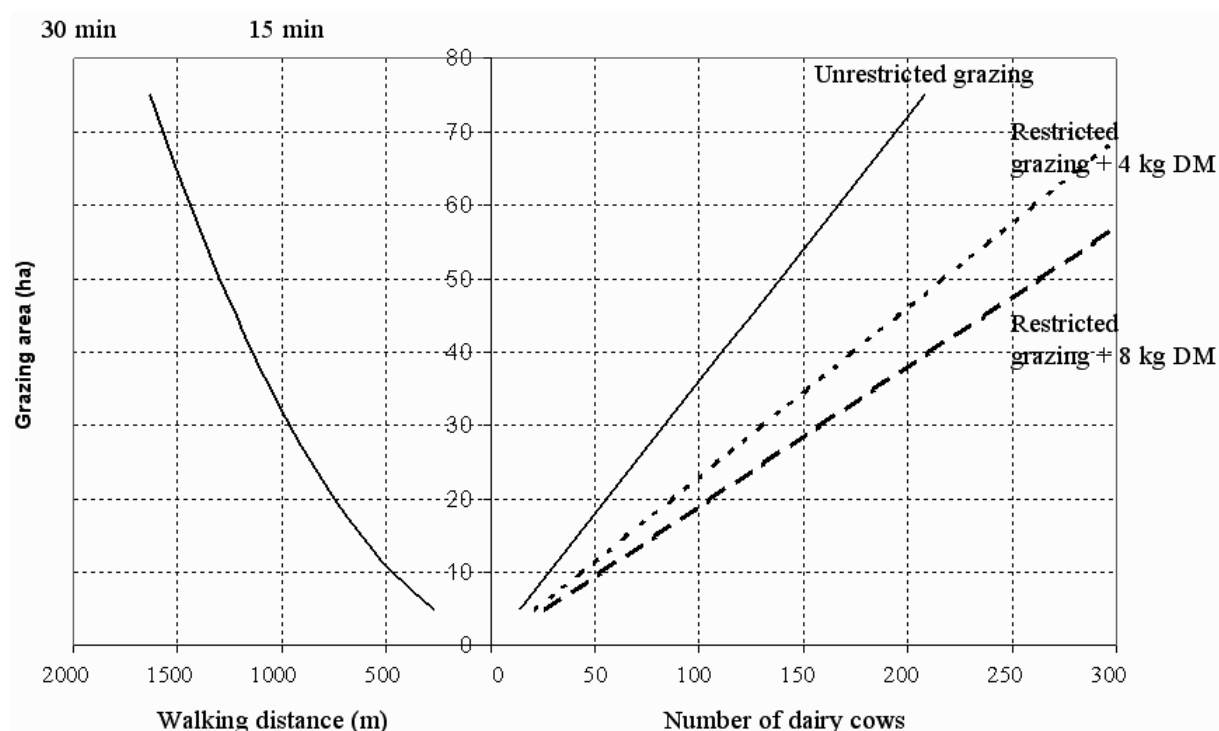


Figure 2. Left: The relationship between the area needed for grazing and the maximum walking distance from paddock to farm buildings. Right: The relationship between herd size and minimum area needed for dairy cows producing 9,000 kg milk yr<sup>-1</sup> and grazing from April till the beginning of October for three systems: unrestricted grazing, restricted grazing (8 hours access) and 4 kg DM d<sup>-1</sup> supplementary feeding and restricted grazing with 8 kg DM d<sup>-1</sup> supplementary feeding.

### Various aspects of grazing

In this paper, systems of grazing and feeding management during the summer are distinguished using the number of grazing hours, since this number determines to what extent the general public notices grazing:

- Unrestricted grazing, grazing day and night
- Restricted grazing, usually only grazing during the day
- No grazing, the animals do not graze

Feed supplementation, with for example concentrates, grass silage or hay, forms an important part of the systems with limited or no access to pasture. When grazing is practised, there are different options to optimize the use of grassland, e.g. variations in paddock size and stocking rate. The choice for a certain system has large effects on grassland production, the animal, the environment, economy and labour and even society. The impact of this choice on various aspects is summarized in Table 2 and explained in this paper.



Table 2. The effect of grazing on various aspects. The score ranges from - - to ++, with ++ signifying that the system concerned scores positive for the point in question, e.g. high health, low losses (Van den Pol-van Dasselaar, 2005).

Viewpoint	Unrestricted grazing	Restricted grazing	No grazing
Grass yield and grass use	-	+	+
Balanced diet	-	+/-	++
Natural behaviour	++	++	+
Animal health	++	+	+/-
Nitrate leaching, N <sub>2</sub> O emission, N losses	-	+	++
P losses	-	+/-	+
Ammonia volatilization	++	+	+/-
Energy use, CH <sub>4</sub> emission	+	-	- -
Fatty acid composition of milk	++	+	+/-
Labour	++	+	+
Economics	+	+	-
Image of dairy farming	++	+	-

### Effect of grazing system on grass production

Grazing affects both grass yield and grass utilization. Grazing has a relatively low gross dry matter production compared to cutting only. This is a result of grass being harvested at a much younger stage. More regeneration periods are needed per year. During grazing losses are incurred due to trampling and deposition of faeces and urine. Conversely, conserving grass gives harvest losses, preservation losses and feeding losses. Unrestricted grazing results in the lowest intake of net energy available for lactation, due to the combination of a relatively low production and relatively large grazing losses.

With a rising milk yield potential, the technical requirements of a properly balanced diet become increasingly important. Because grazing produces relatively large fluctuations in the composition of the diet, the attractiveness of unrestricted grazing declines as the dietary requirements become more demanding.

### Effect of grazing system on animal welfare

Animal health and welfare are important items throughout the entire year and for all animals. Welfare includes aspects that are relatively easy to measure, such as health, and also intangible aspects such as emotions and feelings. Grazing has disadvantages as well as advantages. One important aspect of animal welfare is natural behaviour. This involves the requirements for food, water and rest, and also behavioural needs such as movement, social behaviour, foraging and play. Grazing gives much more scope for natural behaviour compared to conventional cubicle sheds.

Furthermore, grazing reduces the risk of mastitis because the infection pressure of ambient bacteria is lower and there is less probability of the teats being trampled (undamaged teats are less prone to bacterial infection). On the other hand, the sheep head fly (*Hydrotaea irritans*) occurs exclusively outdoors, which means that permanent confinement indoors can avert summer mastitis. But on balance, grazing generally has a positive influence on udder health. Grazing also benefits the claw health of dairy cows. Infectious diseases like foot rot and the disease of Mortellaro are more common in the cowshed, because the infection pressure is higher (Smits *et al.*, 1992; Somers *et al.*, 2005). The relatively hard floor in conventional cubicles can result in wounds and pressure sores on knee and heel joints (Wechsler *et al.*, 2000). The frictional force required for unrestrained locomotion of dairy cattle is minimal 0.6 (Van der Tol *et al.*, 2005). While the frictional force of pasture is higher than 0.8 (Telezhenko *et al.*, 2004), the frictional force of floors in conventional cubicle sheds is in general less than 0.6.

Conversely, grazing results in large fluctuations in the composition of the diet and it makes frequent milking difficult. Both these aspects negatively influence welfare, especially if the cows are very productive. Furthermore, in the field the cows are exposed to the rain and sun and if the temperature exceeds 25 °C, heat stress can occur (Dantzer and Mormède, 1983; Shearer and Beede, 1990). In addition, in the field there is an increased risk of being infected by specific pathogens such as intestinal worms, lungworms and liver fluke. Across-the-fence contact with cows from other farms increases the risk of the transmission of infectious diseases such as infectious bovine rhinotracheitis and bovine virus diarrhoea. However, in practice these risks rarely lead to major problems of animal health. In general, it is easier to prevent the disadvantages of grazing than to remedy the welfare disadvantages of current cubicle stalls.

### **Effect of grazing system on the environment**

Grazing has several effects on the environment, the most obvious being nutrient loss. Less grazing results in reduced mineral losses and thus reduces the imbalance between a farm's mineral inputs and mineral emissions. This is particularly true for nitrogen, but is also important for phosphate. The most important difference between grazing and keeping cows indoors all year is the place where the dung and urine land: some in the pasture, or all in the cowshed. When dung and urine are deposited in the field, a large amount is deposited on a small area where the minerals cannot be used – at least, not in the short term – and thus losses are more likely. Dung and urine collected from the cowshed can be used as fertilizer. This improves the nutrient use efficiency and reduces the need to buy fertilizer, while yields remain the same. Keeping cows indoors all year can reduce a farm's imports of nitrogen by about 50 kg ha<sup>-1</sup> yr<sup>-1</sup> compared to grazing (Van de Ven, 1996). In addition, grazing affects the type of nitrogen loss. During grazing, relatively large amounts of nitrate may be leached (Ryden *et al.*, 1984) and there may be considerable denitrification (Ryden, 1985). Furthermore, there may be relatively large emissions of nitrous oxide (N<sub>2</sub>O) (Velthof and Oenema, 1997). By contrast, collecting dung and urine from the stall and spreading it on the land, as is the case when keeping cows indoor all year, results in more ammonia volatilization. This ammonia volatilization may be partly reduced by adapting the feed strategy (less protein in the ration) (Van Duinkerken *et al.*, 2005). When keeping cows indoors all year, the energy use and hence the CO<sub>2</sub> emissions may also be larger because there is much more use of machinery. The grazing system does not affect methane emissions from grasslands themselves (Van den Pol-van Dasselaar *et al.*, 1999). The larger amount of manure in the slurry pits when keeping cows indoors all year, however, may lead to more methane emissions.

### **Effect of grazing on fatty acid composition of milk**

The grazing system affects the fatty acid composition of milk. Due to grazing, the content of unsaturated fatty acids in milk increases (Elgersma *et al.*, 2003a, 2003b, 2004). Unsaturated fatty acids are believed to be better for human health.

### **Effect of grazing system on economy and labour**

Grazing is, in general, more economically attractive than cutting only. Models indicate that in Ireland early grazing will generate an increased profitability of € 2.70 cow<sup>-1</sup> day<sup>-1</sup> for each extra day at grass, through higher animal performance and lower feed costs (Kennedy *et al.*, 2005). In the Netherlands, the difference between grazing and cutting-only is on average throughout the year € 0.5–€ 2 for every 100 kg milk produced (de Haan *et al.*, 2005). After all, the grazing cow selects, harvests and transports the grass herself and at the same time ensures that manure

and urine are distributed over the field – albeit unevenly. On very intensive farms ( $\geq 20\,000$  kg milk ha<sup>-1</sup>), cutting-only may be economically attractive. The benefits of cutting only on these farms are reduced somewhat by the additional labour needed compared to grazing. The labour input is lowest for unrestricted grazing. Situations of restricted access and no access to pasture require approximately the same labour input. Calculations showed that grazing yields the best returns hr<sup>-1</sup> worked (Van den Pol-van Dasselaar and De Boer, 2007). However, it is not only the number of hours that counts, but also the quality of the labour needed (easy – difficult, light – heavy).

### **Grazing system and society**

The choice for a certain grazing system even has an effect on society. In several regions of Europe, the general public appreciates grazing animals in the landscape. In the Netherlands for example, grazing dairy cows are seen as a national feature. Also, due to grazing, the biodiversity of the landscape increases which is valued by society. Finally, society also associates grazing with animal welfare. Therefore, grazing creates a positive image. The extent to which the general public notices grazing depends on:

- The number of animals
- The area grazed
- The time the animals spend grazing (hours per day, days per year)
- The place of the pasture (next to the motorway or deep in the countryside)
- The moment of grazing (day, night)

### **To graze or not to graze, determining factors**

Farmers have various reasons for choosing a grazing system. In their choice they may incorporate the effect of grazing on grass yield and grass use, but also many other factors like sustainability (economy and environment), animal welfare, and society (Table 2). In some countries legislation is the determining factor.

The importance attached to the various effects of grazing is very personal. For example, what is more important: image of dairy farming or nutrient losses? Moreover, there seems to be conflicting views: grazing has advantages and disadvantages. For most factors the greater the number of hours at pasture, the greater the effect. For some factors the effect is site-specific, e.g. for nitrate leaching. The farm layout is another important site-specific factor which has a big impact on feasibility of alternative systems. It should also be remembered that farm management is an important factor. An individual farmer can have an effect on most of the relevant aspects via his or her management strategy and can thereby reduce or remove the negative effects of a certain grazing system. Table 2 shows that grazing for a limited part of the day scores well on the whole.

However, the question 'to graze or not to graze?' is not that easy to answer. There is another important influencing factor next to the elements already described in this paper. The project 'Koe & Wij' in the Netherlands ([www.koewij.nl](http://www.koewij.nl)) showed clearly that the individual farmer is the main influencing factor. In the end, the personal preference of the farmer determines the grazing system used. Knowledge on the effect of grazing on labour, economy, environment and society is important, but the opinion of the farmer on the effect of grazing on these aspects is affected by personal preferences and experiences. Some farmers have negative experiences with grazing and therefore value all aspects like economy and labour negatively while others with positive experiences do exactly the opposite. Preferences may change with time or during major life events such as expansion of the farm or handover of the farm from parent to child. It may also change as a result of communication with society.

Knowledge transfer to farmers remains necessary. However, since personal preference is an important influencing factor, knowledge transfer alone is not enough to influence the choice of farmers and stop the decline in grazing. Projects to stimulate grazing should focus on personal preferences too. Furthermore, these projects should focus on grazing at large farms since the farm size in northwest Europe will continue to increase. As mentioned before, increased herd size makes grazing management more difficult. Therefore, relatively simple grazing systems should be developed and tested in practice. These systems may not reach the optimum herbage intake, but they will be easy to manage and therefore useful for large farms.

## In conclusion

We expect the trend of decreasing percentages of dairy cattle with unrestricted grazing in Europe to continue. There are economical, practical and personal motives behind the decline in the popularity of grazing. The number of dairy cattle with no access to pasture may be influenced by legislation, knowledge transfer or development of relatively simple grazing systems. In the end, the personal motives of the farmer will answer the question 'to graze or not to graze'.

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We sincerely appreciate the help of Clara Berendonk (Germany), Alex De Vliegher (Belgium), Michael O'Donovan (Ireland), Dave Roberts (UK), Michel Santer (Luxemburg), Karen Søgaard (Denmark) and Eva Spörndly (Sweden) sending information on grazing in their respective countries.

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## **Session 4A**

### **The future of the grassland farmer**



# **Perspectives in dairy production for family farms in different mountain regions of Switzerland**

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## **Abstract**

In the project ‘Mountain Milk’ of the Swiss College of Agriculture a farm-specific strategic and operational planning including a controlling concept based on the balanced scorecard approach was carried out for about fifty dairy farms in five different mountain regions. The goal was integrated sustainability (economic, ecological, social) for the family farms over the next ten years, taking into account the farmer families’ aims, marketing potential and public expectations. Depending on the varying natural and economic conditions the most promising strategies identified differed between regions. In some regions cheese-factories with specialities allow higher milk-prices for small-structured farms with traditional ways of production. In other regions the downward pressure on prices is higher and the farms are strongly forced to realize economies of scale and better efficiency in production.

Keywords: dairy production strategy, mountain areas, optimization, sustainable

## **Introduction**

Most regions of Switzerland are dominated by grassland, especially in mountain areas. The natural and structural conditions are quite different in mountain areas than in the lowlands. The shorter vegetation period limits roughage production and the reduced number of sunny periods impedes the production of high quality hay. Steep surfaces on which mechanization can only be used to a limited extent make roughage conservation the dominant occupation of mountain farmer-families during summer. These restrictions imply comparatively high financial investments in specialized mechanization and infrastructure and impede farm size growth because labour is limited or too expensive and can only be replaced to a limited extent by technical means. Under these difficult conditions, the key question is how to minimize costs while simultaneously building up an improved potential for local added-value of milk production through the marketing of local specialties. Thus a higher milk price can be generated, which is crucial to compensate the very limited cost-reduction potential.

## **Materials and methods**

In the project ‘Mountain Milk’ of the Swiss College of Agriculture a farm-specific strategic planning was carried out for about fifty dairy farms in five different mountain regions of Switzerland. It was based on the classical SWOT analysis approach (optimal combination of strengths and weaknesses with opportunities and threats) which was complemented with operational planning including a controlling concept based on the balanced scorecard approach. The Balanced Scorecard, which is a well established management instrument in economic circles, has the advantage that it directly addresses the four aspects important for economic, ecological and social sustainability, namely learning and growth, internal business processes, customers and finances. The value chain or the description of the farm in the form of processes and influencing factors served as a basis of the analyzing, planning and monitoring process.

## Results and discussion

The predicted results for a time horizon of ten years (2004 to 2013) are promising and motivating for the farmer-families involved. Table 1 shows that the preconditions in 2004 as well as the mid-term perspective differ considerably between project regions. According to the defined development strategies, a competitive dairy production will remain possible in all the project regions and the economic situation of the farms can be improved. The strategies chosen are ambitious but were considered realisable and realistic in the mutual planning process. On average for the 49 farms involved, gross costs per kg of milk produced can be reduced from CHF 1.77 to CHF 1.43 or by almost 20%. Larger and better: these aims are two major approaches for the development of farms. They are propagated by extension services as well by farmers, albeit in varying priority, depending on circumstances. This is one reason for the differing development strategies selected by farms in the different regions of the mountain milk project. Apart from the major approaches 'growth' and 'optimization', which, however, have to be farm-specifically adapted and combined, there is a third approach for mountain farms: 'added value'. This can be achieved by local processing and marketing on the farm or by a local dairy processing SME who is able to offer a better milk price. The 'added value' approach is the basis for the strategy 'improved situation thanks to a higher milk price'. This is in line with the quality leadership strategy of the processing units. The 'growth' approach is categorized as the basis of the 'economies of scale strategy', in line with the cost leadership strategy of the processors. 'Optimization' is considered a prerequisite for any improvement of competitiveness. Both major strategies for dairy production are therefore crucially dependant from the milk buyer and processor and his strategy. Apart from the requirements of the milk buyer, the potential of the region is also important for the choice of the most appropriate dairy production strategy. Last but not least, the individual natural and structural conditions are of crucial importance for the farm-specific choice of the strategy and the implementation of the appropriate farm management. The more difficult and special the natural conditions, the more challenging and special the management will be. Apart from the lower grassland yields, the numerous complex strategic decisions about the most appropriate management are influenced by slope, land consolidation and distances between fields. In principle, for cost reasons, grazing is the most appropriate grassland utilization during the vegetation period. However, the grazing potential is limited through the large requirement for conserved roughage for the winter period which is longer than in the lowlands. Furthermore, grazing with cows might also be restricted by steep slopes, bad land consolidation or large distances from the stable to certain plots. In mountain areas most farms have all types of grassland. The proportion of the different types has an important influence on the appropriate combination of different production systems. Farms with a lot of consolidated but not very flat grassland close to the stable will tend to opt for full grazing with summer accentuated milk production as long as this is not restricted by the milk buyer. Calving is shifted towards the end of winter. This production system is often implemented with the 'growth' approach, leading to lower production costs (Table 2). This approach was chosen by most farms in region 1 (Table 2). The further away and the more parcelled plots are, the more the farm will tend to develop towards winter accentuated production with calving in autumn or early winter. For such farms, grazing is only possible to a limited extent. They therefore try to produce as much high quality conserved roughage as possible and to achieve a somewhat higher milk price through top quality milk in seasons with low milk supply. This 'added value' approach, which was chosen by all the farms in region 4 (Table 1) is leading to reduced growth and higher milk production costs (Table 2).

Mountain farms usually have plots with different slope, parcelling and distance to the stable. In consequence, most farms will end up somewhere between the two basic strategies mentioned above. This makes it very difficult to put forward generally valid recommendations

for the choice of the most appropriate strategy. The problem of very variable land and corresponding management restrictions are often accentuated when farms grow. Solutions are seen in different forms of collaboration between farms. A first step certainly is that farms stop to have both mechanizations for steep and flatter land by changing to collaborative mechanization solutions.

## Conclusions

Every farm has to recognize its opportunities and restrictions when looking for a farm-specific strategy and its targeted technical implementation. In mountain areas it is even more difficult than in the lowlands to define generally valid strategies for the most appropriate milk production. The farm-specific conditions affect the possible production systems, and these in turn have an influence on the milk production strategy. However, the milk production strategy is also influenced by the strategy and the requirements of the milk buyer.

Table 1. Comparison of the structural data of the farms in two of the five regions of the mountain milk project in the initial situation (2004) and in the target situation (2013).

Average altitude Year	Region 1 11 farms 814 m		Region 4 12 farms 1413 m	
	reality 2004	target 2013	reality 2004	target 2013
agricultural surface	25.8 ha	32.7 ha	26.0 ha	26.8 ha
number of cows	21.2	32.5	14.6	16.9
proportion of cows of cattle livestock units	75%	86%	66%	70%
milk yield per cow	6,764 kg	6,721 kg	5,508 kg	5,750 kg
milk produced	143,394 kg	218,419 kg	80,420 kg	97,180 kg
family labour	4,571 h	5,236 h	3,493 h	3,069 h

Table 2. Comparison of the structural and economic data of the farms with the ‘added value’ and ‘growth’ approach in the initial situation (2004) and in the target situation (2013).

Year	Growth approach 21 farms		Added value approach 28 farms	
	reality 2004	target 2013	reality 2004	target 2013
agricultural surface	33.3 ha	36.4 ha	25.3 ha	27.0 ha
number of cows	23.4	29.0	17.9	22.8
milk yield per cow	6,108 kg	6,365 kg	5,986 kg	6,221 kg
milk produced	142,930 kg	184,577 kg	107,153 kg	141,839 kg
family labour	5,401 h	5,540 h	4,653 h	4,534 h
Gross costs per kilo of milk produced (CHF)	1.60	1.32	1.90	1.52
Performance per kilo of milk produced (CHF)	1.41	1.26	1.63	1.37

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## Mobile milking robot offers new grazing concept

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### Abstract

Grazing of high-producing large dairy herds is increasingly under pressure in Europe. Due to increasing farm size, fewer enterprises possess enough land around the farm buildings to offer sufficient grazing. This limits access to the numerous beneficial effects of grazing which include improved animal health, reproduction, product quality and crop rotations.

Automatic milking systems (AMS), when made mobile, give the possibility of milking cows in the field without additional labour. Modern technologies for surveillance and livestock control facilitates the management, even if miles away from the barn. During winter, mobile AMS can service animals in the barn.

A prototype mobile milking robot was built as part of an innovation research programme, with collaboration between Århus University, industry, an organic dairy farmer and an organic crop grower. The prototype was tested in a herd of 90 cows grazing 15 km from the farm buildings, in the summer of 2007. The promising results suggest that this new management system will allow grazing to take place away from the farm and allow crop farmers to include grazed clover-grass pastures in their rotation. Further documentation is necessary before marketing the concept. Cow traffic in the field, animal welfare, production potential, and grazing management are scheduled to be monitored and recorded in the season of 2008.

Keywords: dairy cows, grazing concept, mobile AMS

### Introduction

Reduction of grazing for highly productive dairy cows and large herds is noticeable in severable countries like Denmark, the Netherlands and Germany (van der Poll, 2005). This gives concern amongst consumers who perceive a decline in welfare of housed animals (Somers *et al.*, 2005) and in product quality (Anonymous, 2006). In addition, the impact of zero grazing on the system's energy balance and economy are unsure. With rising energy prices the cut-and-carry system for feeding roughage in the barn is becoming more expensive. Automatic milking is gaining market share, not in the least because of reduction in physical labour needs and the farmer having increased flexibility. After having overcome technical difficulties, the main concern relating to Automatic Milking Systems (AMS) is grazing management (Oudshoorn *et al.*, 2007). Grazing reduces milking frequency (Ketelaar-de Lauwere *et al.*, 1999) and increases the time taken to fetch cows for milking. Decreased milking frequency can be due to the synchronized behaviour of the herd (Munksgaard and Søndergaard, 2004), and the preference of cows to be out on the grass.

In addition, the lack of adequate areas of pasture adjacent to the barns, due to increasing herd size, can be a problem (Oudshoorn and de Boer, 2005). These aspects were addressed in the concept for mobile robotic milking, developed in cooperation with S.A.Christensen A/S and practical farmers. The portability makes it possible to utilize allocated pasture away from the barn but still use the expensive equipment in the barn during winter. Verification and documenting the concept, by building and using the mobile milking robot on a farm, was initialized with help from innovation funding in Denmark.

## Materials and methods

A technical design of a mobile unit which could milk dairy cows in the field was made in 2003 ([www.automaticmilking.dk](http://www.automaticmilking.dk)). Challenges for this design were its mobility because of the weight, and compactness, as a standard container was preferred for the frame (Figure 1). A prototype was constructed in the spring of 2007; it uses a two-unit milking robot and weighs around 12 tonnes. It was placed in a 30 ha pasture, besides a dirt road, to service a herd of 90 mostly RDM cows. The water supply was provided by pipe, and the electricity supply for milking and milk refrigeration by generator. The unit was placed in the pasture at the beginning of June 2007. The herd had not previously been milked automatically. An automatic gate system regulated the movement of cows from one paddock to the next. To start training the herd to automatic milking, the grazing system was made as simple as possible; a regulated permanent grazing system, with two pastures. The admittance to respective pastures by separation gate was controlled by a timer and central processor (CP), which identified cows entering the robotic unit. The CP only allowed cows out to new pasture if they were not due to be milked within one hour.

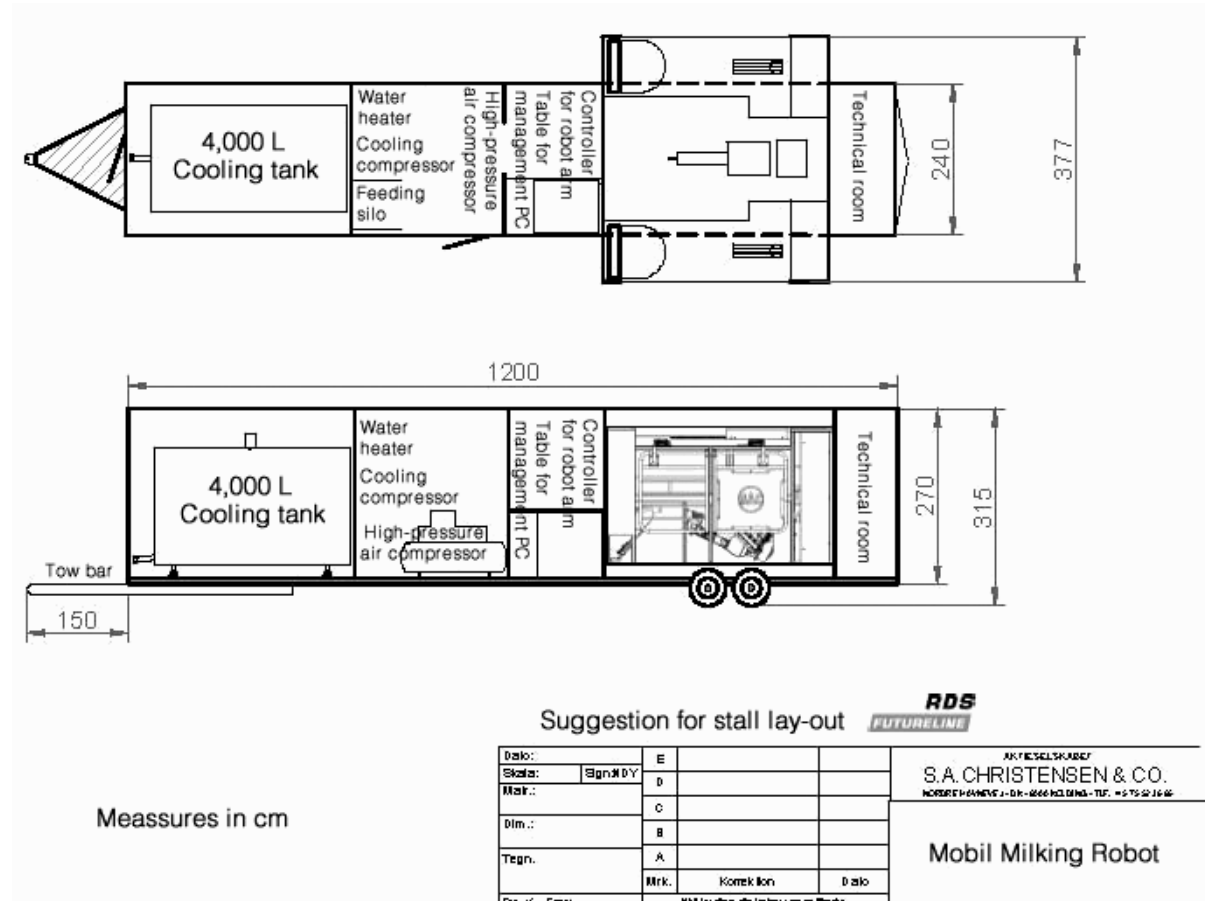


Figure 1. Industrial design for mobile milking robot.

## Results and discussion

The adaptation to robotic milking proceeded satisfactorily. After 1 week, 90% of the cows could be milked automatically and after 4 weeks 90% of the herd entered the milking units by themselves. However, the number of cows being milked at least twice a day, without human interference, fluctuated greatly. During the 'learning' process, adjustments were made to the catching dock, due in part to soil damage at gateways as a result of the high rainfall frequency



during summer and the lack of farm management experience with in-field milking. This frustrated the cows, as they are very much slaves to routine.

Initial results from two months of practice with the AMS in the field were compared to the same months the year before, and they showed similar results in hygiene and milk quality (Table 1). The cows received 2 SFU (Scandinavian Feed Units) as concentrate per day; grazing was unlimited in both years, and was estimated to exceed 11 SFU per day. An average of 1.8 milkings per cow was recorded in July/August, and cows visited the milking site individually, mostly at daytime.

Table 1. Milk quality parameters as averaged for two comparable months in 2006 (conventional milking system) and 2007 (automatic milking system).

	Jul/Aug 2006	sd	Jul/Aug 2007	sd
Herd size	68		90	
kg ECM <sup>1</sup> /cow	5900		5200	
SCC <sup>2</sup> (1000 per MI)	388	98	376	60
				122
TBC <sup>3</sup> (1000 per MI)	10	17	387	0
				0.0
Fat %	3.89	0.06	3.89	9
				0.0
Protein%	3.3	0.07	3.3	5
Urea (Mmol L <sup>-1</sup> )	6.7	0.7	5.7	1.3
Spores of anaerobes L <sup>-1</sup>	145	271	55	45

<sup>1</sup>ECM: Energy Corrected Milk, <sup>2</sup>Somatic Cell Count, <sup>3</sup>Total Bacteria Count.

## Conclusions

The successful start of the mobile in-field milking system will be followed up by extensive registration of data in the grazing seasons of 2008 and 2009. Technical design and functioning of the AMS was capable of delivering good results in an outside environment. Around-AMS logistics and design of the catching dock around the milking robot have to be improved.

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# Local participation in seminatural grasslands maintenance: lessons from Sweden

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## Abstract

This paper concerns local participation in the protection and maintenance of biological and cultural values in seminatural grasslands in Sweden. The values of seminatural grasslands are dependent on continuous management and animal husbandry, why farmers play a crucial role in the maintenance. The study presented aims at exploring prerequisites for increased local involvement. Two cases, where the local involvement is considered successful have been investigated; Southern Öland, where community involvement in seminatural grassland management has been experienced in Life-projects and in the process of becoming a UNESCO World heritage site, and Mälärhagar, an integrated restoration and beef production project carried out in close collaboration with farmers. The results are discussed in terms of good and bad experiences as well as difficulties to handle. The findings show that trust, communication and local influence are vital ingredients in participatory approaches. There are, however, problematic collisions between directives on nature conservation and directives on public participation.

Keywords: local participation, seminatural grasslands, landscape management and planning, nature conservation

## Introduction

How should the management of certain qualities in seminatural grasslands be organized in terms of strategic planning and decision-making? While nature resource management has traditionally been characterized by top-down perspectives, the buzzword these days is participation, as is seen in its influence upon policies concerning the cultural landscape (Selman, 2004; Stenseke, 2006). However, the meaning(s) and implications of local participation have not yet been thoroughly investigated. The objective of this paper is to enhance knowledge about the prerequisites for local participation in the planning and management of cultural landscapes, and to reveal critical aspects that have to be considered when introducing participatory strategies into landscape management and planning. Focus is set upon semi-natural pasture management and the collaboration between farmers and official executives. Some Swedish experiences are presented and analysed; Southern Öland and 'Mälärhagar'. Both of them have been recognized as good examples of landscape management that includes local involvement (Stenseke, forthcoming). In Sweden, the fact that the greatest threats to values identified in the cultural landscape are the decreasing number of farmers, and the afforestation of fields and pastures, has fuelled an awareness within the Swedish political and administrative system of the necessity to construct landscape management strategies that are perceived by farmers and would-be farmers as being both positive and stimulating. Most of the agricultural land in the country is privately owned, and contains common goods such as biodiversity, cultural heritage and recreational space. These values are mostly maintained in the context of animal husbandry and grazing. Southern Öland holds vast areas of seminatural pastures, extremely rich in biodiversity. In 2000, the agricultural landscape of southern Öland was included in the UNESCO World Heritage List. Farmers played an important role in the nomination process, and have a significant stake in



governing the maintenance of the area. The case of Mälarhagar concerns a number of valuable grasslands and farms spread out over a region. The shores of Lake Mälaren (In Swedish: Mälarhagar) have traditionally been used as grazing lands. 'Mälarhagar' was initiated as an integrated biodiversity restoration – beef production – outdoor recreation project. As a result of the project, 25 farmers started a business for a joint trading of beef and registered the trademark 'Mälarhagskött' (Mälar pasture beef).

## **Materials and methods**

Data from the two cases have been collected mainly through interviews with stakeholders, most of them farmers, but also executives at various authorities and other organizations. Furthermore, inventories, planning documents, protocols and general statistics have been examined. The ambition has been to get a broad picture of processes, structures and events. Respectability, legitimacy and robustness have been the criteria that have governed the data collection by motivating an investigation of the course of events from the perspective of various actors; to what extent can the processes be considered as fair, and are there indications that the solutions launched will be long-term?

## **Lessons from Sweden**

What do we learn from Southern Öland and Mälarhagar about the prospects for local participation? Features such as trust and respect, structures for local influence, common understanding, market potentials, time and money all appear as success ingredients in these Swedish cases, while inflexible top-down directives and inefficient attention to the conditions for co-management prove to be hindrances in establishing a participatory approach in landscape management and planning. The case studies also point out a number of difficulties one needs to be aware of and to handle when initiating local participation; the history of the relations between the local area and the regulating authorities, information barriers, local power structures and the ever changing character of reality, implying the need for capacities to handle altered preconditions. Though having a local focus, the results illuminate some general issues about prerequisites and critical aspects for local participation in the planning and management of the cultural landscape. One issue is the collision between constitutional rules in a specific piece of landscape. Nature conservation goals aiming at enhancing biodiversity, implemented using powerful measures, stand in contrast with the principle of local involvement and public participation. As landscape policies and landscape management strategies have developed during an era of powerful and mandatory nature conservation regulations, the competence within the administration is much more advanced concerning conservation issues than it is about communicative and participatory issues. It is, however, not a proper solution to construct less detailed regulations, giving room for the negotiation and consideration of local interests, as that implies a risk that biodiversity and cultural heritage values will not be given the necessary attention. This points towards a need for transparency, making clear what qualities are regarded as national and international interests, and why, and what could be locally decided. Another critical aspect concerns the subject for collaboration. In the planning and management of the cultural landscape, there is a multitude of considerations to take into account, greatly varying in scale and focus. One important observation with implications for local participation is that there is no clear correlation between scale and requests for local influence. For the farmers, and also for the local municipality, it was seen to be important that the management of the cultural landscape values is considered and discussed within the broad contexts of agriculture and rural development. Thus, to the local stakeholders it is not satisfying if they are invited to co-operate only on the operational level, when it comes to activities in the physical landscape. Instead, they also

want to participate in defining the institutional arrangements for landscape management. When landscape management is discussed within a wider context, as was done in various formal arenas on Öland, and in seminars for farmers in Mälärhagar, it means a better coherency with local stakeholder perspectives. Consequently, increased local participation demands a development from sector-bound planning towards a more integrated landscape perspective in the political and administrative system as well. A third aspect of vital importance relates to democracy. In co-management of the cultural landscape on Southern Öland and in Mälärhagar, farmers were the clearly dominant local stakeholders. It could be questioned whether the powerful role of farmers is reasonable, as they constitute a minority of the local population. Since the issue mainly concerns farmers' private land and their activities on their properties, it could be regarded as a fair argument that they should have a strong influence. On the other hand, the transformation of the cultural landscape from a production resource to a product in itself motivates reconsideration of who are the stakeholders and who should have a say. Such a development is likely to bring changes in what constitutes property rights, even implying consequences for future agriculture and forestry. When decentralising power, the question of 'to whom or to which group of people' needs to be asked. If democratic aspects are not considered, local participation might turn out to be less democratic than top-down power structures.

### Concluding remarks

There are indeed some good lessons to learn about local participation from landscape management in Southern Öland and Mälärhagar. However, it has to be stressed that to fully copy the good examples of collaboration is not a guarantee for success. In both investigated areas, co-management is to be regarded as an evolutionary product, including networks of relations that are developing over time. The challenge for the political and administrative system is to design fair landscape management policies that permit local solutions. Place specific conditions such as the existence and numbers of active farmers, the range of stakeholders, the possibilities of getting agri-environmental payments, the existence of potential consumers of local products within a reasonable distance etc. are of vital importance for what kind of collaboration could be developed and established. Many parts of Sweden are suffering from depopulation and in quite a few villages there are no active farms any longer. Similar trends can also be seen in many other parts of Europe. This means a different set of prerequisites as the fields and pastures have to be managed by farmers from other places, who might not be the owners but are renting the land. Another conclusion is that an adjustment of the idea of what competences are necessary for executives working with landscape management and planning is needed. The contemporary trend in nature resource management from nature protection to social ecological resilience and governance (Folke *et al.*, 2003), implies that skills in communication and collaboration are of vital importance. It is indeed necessary to get away from the understanding that nature conservation is about biology, and instead to recognize the fact that to a significant degree it is about interactions with people.

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# Biogas yields from grassland

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## Abstract

Biogas production is a suitable way of energetic use of surplus grassland. The influence of grass species, cutting periods, intensity of grassland use, and ensiling additives on biogas and methane yields was investigated in combined field and laboratory tests. Methane yields of grassland biomass vary in a wide range, mainly depending on the intensity of grassland use. As a rule, an intensive management with an early first cut and several cuts per year leads to both high substrate-specific and area-specific methane yields and vice versa. The influence of grass species and ensiling additives on substrate-specific methane yields seems to be secondary. Further systematic research on optimal grassland management for biogas production and on environmental and socio-economic impacts is required.

Keywords: grassland, biomass, bioenergy, biogas, methane yields

## Introduction

Grasslands play an important role in European agriculture, covering a total of more than 69 million ha in the EU-27, i.e. 36% of the utilized agricultural area (FAOSTAT). It is estimated that the theoretical grassland potential for energy and materials in the EU-27 amounts to 9.2-14.9 million ha (Prochnow *et al.*, 2007). Grassland biomass is principally suitable for manifold ways of energetic use (biogas, combustion or biofuel) and material use (e.g. fibre or chemicals). Among these, biogas production currently is the process most commonly implemented in practice. Biogas and methane yields from grassland depend on various factors such as vegetation type, management and preservation practices. Knowledge of the effects of these factors is required to optimize the use of grass for biogas production. The aim of this paper is to give an overview on several works of the authors on biogas yields from grassland.

## Materials and methods

The influence of grass species, cutting periods, intensity of grassland use, and ensiling additives, on biogas and methane yields was investigated (Table 1). Grass samples were taken from field experiments and digested in laboratory tests (for details see references in Table 1).

## Results and discussion

Grass species: Substrate-specific methane yields of the grass species investigated count for 310-360 l<sub>N</sub> kg<sup>-1</sup> VS (l<sub>N</sub> – Normal Litre, VS - Volatile Solids), showing no clear differences between the species (Mähnert *et al.*, 2005). Other authors present similar results, obtaining biogas yields of 480-540 l<sub>N</sub> kg<sup>-1</sup> VS (Baserga and Egger, 1997) or methane yields of 300-350 l kg<sup>-1</sup> VS for different grass species and varieties (Kaiser and Gronauer, 2007). The influence of grass species on substrate-specific methane yields seems to be secondary.

Table 1. Test series on biogas yields from grassland.

Test series	Remarks	Reference
Grass species: grass from pure stands of perennial ryegrass ( <i>Lolium perenne</i> ), cocksfoot ( <i>Dactylis glomerata</i> ), meadow foxtail ( <i>Alopecurus pratense</i> )	first cut, mid May 2001, samples fresh and ensiled, sand and fenland sites in northeast Germany	Mähnert <i>et al.</i> , 2005
Cutting period: grass from landscape management, samples taken monthly from June to March, always first cut, 2001-2004	fresh samples, meadow foxtail grassland ( <i>Alopecuretum pratensis</i> association) on a fenland site in northeast Germany	Prochnow <i>et al.</i> , 2005
Intensity of use: grass from a mountainous site, extensively used, and a valley site, intensively used	one two four cuts per year at varying stages of vegetation, samples fresh and ensiled from meadows in the Austrian Alps, 2004	Amon <i>et al.</i> , 2005
Ensiling additives: starter cultures, homofermentative ( <i>L. plantarum</i> , <i>L. rhamnosus</i> ) and heterofermentative ( <i>P. pentasaceus</i> , <i>L. brevis</i> , <i>L. buchnerie</i> , <i>L.</i> ) inoculants	rye grass ( <i>Lolium multiflorum</i> L.), properly managed permanent grassland in northeast Germany, first cut mid May, second cut early July 2005, samples fresh and ensiled	Idler <i>et al.</i> , 2007

Cutting periods: The results obtained show decreasing substrate-specific methane yields with delayed cutting periods. Methane yields decline linearly from 298 l<sub>N</sub> kg<sup>-1</sup> VS in June to 155 l<sub>N</sub> kg<sup>-1</sup> VS in February (Figure 1) (Prochnow *et al.*, 2005). The increasing content of crude fibre restricts the maximum biogas production potential since crude fibre consists mainly of hemicelluloses and lignin, both described as hardly bio-degradable under anaerobic conditions (El Bassam, 1998). Area-specific methane yields obtained reach a maximum of 1,700 m<sup>3</sup> ha<sup>-1</sup> in August/September.

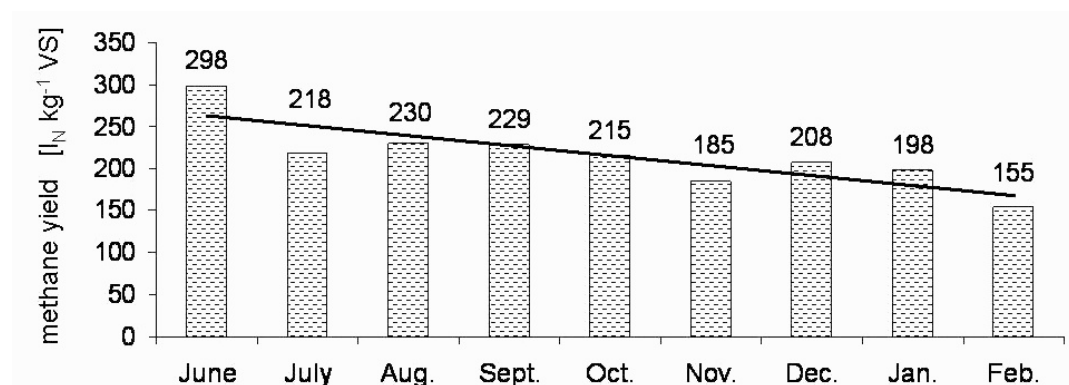


Figure 1. Seasonal pattern of substrate-specific methane yields from landscape management grass (mean values of three years and trend line).

Intensity of use: In the Austrian Alps intensive management of a valley grassland site with three to four cuts per year yields 2,746-3,459 m<sup>3</sup> ha<sup>-1</sup> of methane. Extensive use of a mountainous site with one to three cuts results in methane yields of 649-1,108 m<sup>3</sup> ha<sup>-1</sup>. (Figure 2) (Amon *et al.*, 2005). Substrate-specific methane yields decrease with advancing stages of vegetation, counting for 221-362 l<sub>N</sub> kg<sup>-1</sup> VS during stem elongation and inflorescence emerged, for 171 l<sub>N</sub> kg<sup>-1</sup> VS during anthesis and for 153 l<sub>N</sub> kg<sup>-1</sup> VS in August. Period of first cut also dominates area-specific methane yields of the entire year, even for intensive management with several cuts per year.

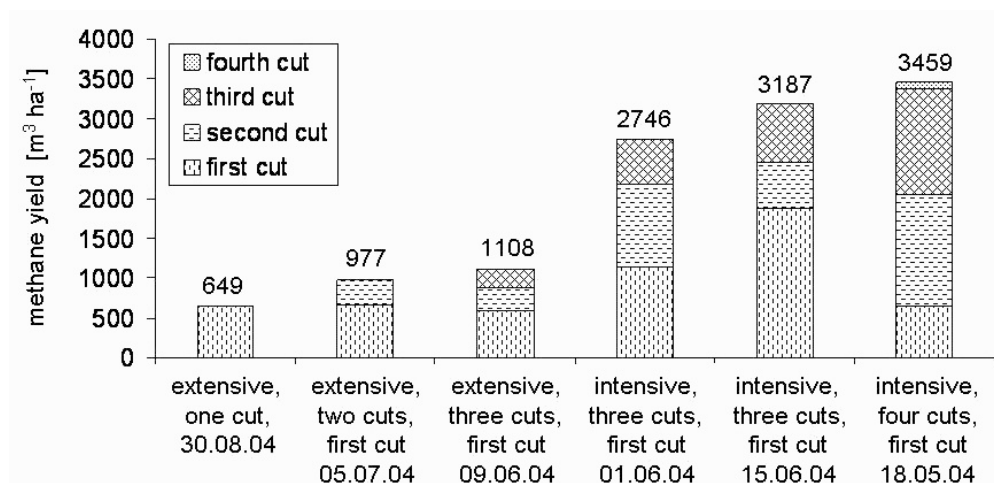


Figure 2. Area-specific methane yields of grassland with different intensity of use in the Austrian Alps.

Ensiling additives: Research on preservation and storage of grass as feedstock for anaerobic digestion is still at the emerging stage. First results indicate various effects of additives on the substrate-specific methane yield of silages prepared from cuttings of permanent grassland (Idler *et al.*, 2007). Hence, the main focus of current experiments is the interrelation of additive treatment and extent of storage losses. Finally, effectiveness of additive usage for preparing silage as feedstock for anaerobic digestion has to be quantified by intended economic assessments.

## Conclusions

Methane yields of grassland biomass vary in a wide range, mainly depending on the intensity of grassland use. As a rule, an intensive management with an early first cut and several cuts per year leads to both high substrate-specific and area-specific methane yields and vice versa. Further systematic research on optimal grassland management for biogas production and environmental and socio-economic impacts is required.

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# The boom in biomass production – a challenge for grassland biodiversity?

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## Abstract

The current boom in biomass production in order to mitigate climate change massively affects agricultural production in Germany. It causes a large-area intensification and loss of grassland, mostly in favour of maize. This development is a great threat for the diversity of grassland communities. It is of utmost importance for the scientific community and political stakeholders to tackle these challenges and to develop strategies for the use of grassland which comply with biodiversity targets and help to mitigate and adapt to climate change.

Keywords: biomass production, grasslands, high nature value areas, biodiversity

## Introduction

The current boom in biomass production throughout Europe is caused by efforts to reduce climate change through the use of renewable vegetable raw material instead of fossil fuels. In Germany, the Renewable Energy Sources Act from 2004 is the most important law for the extent and practice of biomass production. Since its coming into force, both the number of biogas plants and the dimension of electricity generation have increased considerably (Fachverband Biogas, 2007). In 2007 two million hectares of arable land has been used for the production of vegetatal raw materials in Germany. A sharp rise from 2003 to 2004 can be noticed (Fachagentur nachwachsende Rohstoffe, 2007).

Two political processes play a key role in future developments in biomass production. Firstly, the increasing political importance of mitigating climate change. Secondly, and in relation to the former, a new strategy for an integrated European climate and energy policy is currently under development. In early 2007, the European Council has established a framework for the further development of a European joint approach in this respect. As a consequence, it is to be expected that the sharp increase in transforming farmland for biomass production will continue. In particular, the ambitious and binding EU-target to increase the share of renewable energies in primary energy consumption to 20% by 2020 will influence land use considerably (BMU, 2007).

A forecast of the Wuppertal-Institut (2006) presumes for the year 2030 a 3.0 million hectare acreage for the production of vegetal raw materials for Germany. The potential for biomass-production on grassland and cropland is estimated between 14 and 23% of the entire agricultural area, which is between 2.47 and 3.94 million hectares (Fritsche *et al.*, 2004).

Against this background it is the aim of the text to consider, on the basis of literature review and unpublished expert informations, if there is an impact of these changes on grassland, especially on grassland biodiversity. On this basis the need of scientific programmes and political strategies is deduced. This paper wants to arouse both, scientific community and political stakeholders to discuss and develop strategies for the use of grassland which comply with biodiversity targets and help to mitigate and adapt to climate change.

The German Federal Agency for Nature Conservation is currently undertaking a research project developing 'Nature conservation standards for Biomass production' in order to tackle the above mentioned challenges. First results will be presented at the conference in Uppsala.

## The influence of biomass production on grassland in Germany

The increase in biomass production for energy has a substantial impact on agricultural areas, in addition (and in competition) to nature conservation as well as the production of food and fodder. There is evidence to hypothesize that this impact is not only concerning arable land, but also grassland. But most of the research regarding changes in land use is not focused on grassland and grassland biodiversity. To verify or falsify the hypothesis and to close this gap of knowledge the literature review is supplemented by unpublished expert information, observed by several conservationists in Germany.

Reports from numerous European regions state that an increase in biomass production does not only affect cash crop farming, but also has increasing direct and indirect impacts on grasslands. In Germany, 30% of the biogas-plants use grass-silage as a co-ferment, the percentage of grass as a co-ferment is about 5%. This usage has a direct effect on biodiversity, because the best grasslands for high-quality silage for biogas-plants are species-poor, *Lolium*-dominated and cut in a small timeframe in spring before the inflorescences are visible (Hochberg *et al.*, 2007). The connection between usage of grassland for biogas and a higher number of cuttings, which is an important component for the intensity of grassland cultivation, is documented by Agroplan (2006) in a survey of farmers with biogas plants. 17.5% of the answers reported a modification in the number of cuttings since the construction of the plant. Nearly 43% of them make one additional cut, 25.0% two and 3.6% three more cuts. However, 21.4% of the farmers make one or two cuts fewer (7.1% not specified). Even former species-rich grassland is now used as co-substrate, with the well-known negative effects on biodiversity of more cuttings, increased fertilization and an earlier first cut.

Indirect effects are also causing problems. Due to the high prices for agricultural products, there is an ongoing conversion of grassland to arable land. This conversion is mainly going hand in hand with the cultivation of maize, which has even been monitored in Natura 2000 sites. It is reported that regions with a high density of biogas plants are drowning in maize (Koop, 2007). This development will be the biggest threat for grassland and its biodiversity in the next years. But not only conservationists observe this change in land-use. It affects also public and touristic interests in cultural landscapes.

The development is accompanied by a loss of pasture areas caused by problems related to livestock farming, where farmers cannot afford the increasing rental prices for land (Kraft, 2007). All these factors cause negative effects on the biodiversity of grassland communities. Although biodiversity of extensive grassland which has been intensified or converted into cropland is restorable, this can only be achieved over long periods with high maintenance. Therefore, the preservation of extensive grassland should be the first priority (Rösch *et al.*, 2007). A chance for preservation of grassland biodiversity is to replace traditional ways of usage which are no longer in demand, e.g. forage production, with the use of grass for energy production. This concept may avoid fallows especially on marginal habitats, and would save biodiversity as a consequence (Rösch *et al.*, 2007).

## Conclusions

From the nature conservation side it is important to meet these challenges and to develop a strategy to stop the increasing loss of grassland biodiversity. Elements of this strategy could be:

- to revise legal instruments such as Cross Compliance and good agricultural practice in the light of massive change of land use as consequence of biomass production. It will be important to stop the intensification of grassland usage and the conversion of grasslands into arable land for biomass production, in particular, but not exclusively within protected areas;



- to further develop an ecologically qualified substitution system for the discontinued set aside arable land schemes within the Common Agricultural Policy (CAP);
- to implement national and international certification standards for biomass production which include both biodiversity aspects and greenhouse gas (GHG) balances;
- to strengthen the 'second pillar' of the European CAP in order to better finance and enable nature protection and landscape conservation, for example by pasturing and harvesting meadows especially in high nature value farmland areas, which play a decisive role for the species richness of grassland communities;
- to finance research programs which develop optimised grassland cultivation systems (e.g. seed-mixtures) to enhance grassland yield for the use in biogas facilities as well as optimising systems increasing grassland biodiversity, including agro-forests schemes;
- to finance research programs which compare different grassland cultivation systems (high and low intensity as well as high and low biodiversity) with regards to GHG-balances, and to compare organic and conventional grassland farming concerning the effects on grassland biodiversity and their GHG balance;
- to develop different systems of grassland cultivation that comply with biodiversity targets and help to mitigate and adapt to climate change.

As the CAP payments are also justified for the maintenance and improvement of biodiversity and to address the overall EU-target of halting the loss of biodiversity by 2010, it is of utmost importance for the scientific community and political stakeholders to tackle these challenges.

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## **Agroforestry systems: from tradition to future sustainable land use**

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### **Abstract**

Agroforestry has been identified as sustainable land use management by UN in the Agenda 21. Recently, the EU included direct payments for establishment of agroforestry systems in the last Rural Development Directive. There are different types of agroforestry systems; the most important in Europe are silvoarable (including forage production) and silvopastoral systems. Agroforestry systems are currently used from northern (reindeer husbandry) to southern (dehesa, montados) latitudes of Europe. The implementation of agroforestry systems must take into account the tree component in order to enhance synergies between pasture and tree components. Agroforestry systems have also been described as a way to enhance biodiversity and to reduce nitrate leaching contamination and promote carbon sequestration. This paper reviews the main types of agroforestry systems related to animal husbandry in Europe and the current productive, environmental and social benefits they have.

### **Introduction**

Grazing animals have used forest areas since historic times. Wild herbivores are, and were, part of the forest ecosystem (Rois *et al.*, 2006). They modified plant relationships in the forest, depending on stocking rate and grazing pressure; these in turn depended on periods of feed shortage linked to inter-annual variations in climate. As human beings became sedentary, wild animals were replaced by domestic animals and the influence of animals on forest areas became more important, and vegetation was modified and co-evolved as a result of their impact. Man acted as a moderator of this relationship and learned to use forest resources to feed different types of animals like cattle, goats, sheep or pigs in a sustainable way. Continuous and very intensive grazing can reduce tree density, and finally destroy the woody component due to the intake of the woody plants and the tree saplings. In Europe, intensive agriculture has been promoted since the 18th century, and the dependence of the agricultural areas on forest areas (nutrient cycling) was reduced due to fertilization and liming practices. These practices were specially applied in most of the Atlantic and Continental biogeographic region of Europe, where forest area was drastically reduced, with the exception of mountain areas. The need for improvement of efficient use of resources made the deforestation less important in Mediterranean or Alpine areas of Europe. Intensive management systems of those Atlantic and continental areas of Europe has caused environmental and ecological problems (eutrophication, biodiversity loss) which could be partially solved through the implementation of agroforestry systems, an option which was promoted in the last European Rural Development Regulation (EU, 2006).

### **Agroforestry systems in Europe**

Agroforestry systems can be defined as the combination of a woody component with an agronomic crop. Agroforestry systems are extensively applied in tropical areas due to the lower impact of human beings on natural ecosystems. Agroforestry systems combine different agroforestry practices (Nair, 1993). European types of agroforestry practice are silvoarable, forest farming, riparian buffer strips, silvopasture, improved fallow and multipurpose trees (Mosquera-Losada *et al.*, 2007a). The most important agroforestry practices in Europe are

silvopasture and silvoarable (Eichhorn *et al.*, 2006), but examples of the others can be easily found (Mosquera-Losada *et al.*, 2007a). Silvoarable and silvopasture practices are usually linked to domestic animal management because in the first case crops are linked to feed animals during shortage periods (hay, silage) and in the second case animals are a component of the agroforestry practice. Both can be easily found in the Mediterranean area (Eichhorn *et al.*, 2007; Mosquera-Losada *et al.*, 2007a), but they can provide some benefits in other areas like Continental and Atlantic biogeographic regions of Europe. Silvoarable agroforestry practice is defined as obtaining of annual or perennial crops in between widely spaced trees. Silvoarable practices can be split into *alley cropping*, when trees are grouped in rows, *scattered trees* (trees established at a low density, not in rows) and *line belts* (hedgerows, shelterbelt, windbreaks and forest belts) when trees are distributed around farms of plots. Silvopasture, the combination of trees and animal production, can be divided into *forest or woodland grazing* (high density stands, natural forests) and *open forest trees* (low density stand, recently afforested or reforested areas), and animals are part of the component.

### **Silvopasture and silvoarable agroforestry practices and animal production**

Domestic herbivorous animal production system designs usually include grazing and arable lands, which rotate in order to provide fresh pasture and stored food to feed animals during the shortage periods. The proportion of land allocated to grazing and to arable crop production (mainly maize (*Zea mays* L.), but also Italian ryegrass (*Lolium multiflorum* Lam), rye (*Secale cereale* L.)) depends on the climatic conditions of the area associated to droughts or cold. Most intensive animal production systems based on pasture have been described as part of the diffuse contamination focus (nitrate, biodiversity loss) if no adequate management is made. The introduction of trees at a low density in grasslands or croplands could provide some advantages related to productivity and environmental issues. Regarding productivity, the land value is increased due to the tree growth that could be sold as a high quality wood. This kind of system is successfully implemented in New Zealand, and has demonstrated good profitability at an experimental level in different parts of Europe (Rigueiro-Rodríguez *et al.*, 2007). On the other hand, they fulfil some of the main priorities highlighted in the last Rural Development Regulation as they enhance animal welfare, being in line with the production of healthy animal derived products of high quality. In fact, this could be used to produce animal labelled products linked to farm tourism (Pardini, 2007). Tree presence in grassland can also provide benefits from an environmental point of view, mostly related to biodiversity preservation, efficiency on nutrient resource use and carbon sequestration enhancement. Biodiversity enhancement is attained through the heterogeneity created by the presence of the tree in the land which creates patches where light inputs are modified (under canopy and open sward) and create different microclimates in the sward depending on tree distribution and age (Mosquera-Losada *et al.*, 2005). The animal component also modifies sward vegetation relationship due to the selection of plants, trampling and faeces distribution; all these parameters are related to the stocking rate. But also, agroforestry can help to reduce habitat fragmentation (one of the main biodiversity problems in some Atlantic regions) and to preserve domestic animal breeds in risk of extinction more adapted to marginal areas. In the Mediterranean areas, biodiversity preservation is promoted due to the reduction of fire risk caused by silvopasture practices (Etienne *et al.*, 1996; Rigueiro-Rodríguez *et al.*, 2007). Tree presence in grassland, allows tree roots to grow below the grassland rhizosphere, and utilize nutrients not used by grassland roots, promoting tree growth and, at the same time, avoid nutrient leaching and associated water contamination. On the other hand, the development of tree rhizosphere below tree grassland roots enhances carbon sequestration, when compared with land use exclusively for grassland.

## Tree and pasture management

Tree and pasture management should be based on synergies promotion. Tree cover is an important factor because it modifies light input to grassland. Tree cover depends on age and tree density and silvicultural practices should be carried out to promote light input (Mosquera-Losada *et al.*, 2005) like thinning or pruning. Final tree density is usually low (25-50 or 100 trees ha<sup>-1</sup>), but depends on tree crown type. An adequate selection of grass species should be made; e.g. *Dactylis glomerata* has demonstrated good resistance to shading conditions (Mosquera-Losada *et al.*, 2007b). Animal type is also an important issue in order to establish silvopasture practices. Tree damage should be avoided, as it reduces long-term profitability of the agroforestry system, and their environmental benefits. Some animals, like goats, cause more tree damage than sheep, but the impact of the damage depends on animal stocking rate, grass availability and the animal breed involved in the practice. Tree protectors should be used to protect trees from animal damage, but for a short period of the whole stand-life period if an even stand is used, i.e. coming from afforestation or reforestation. The main difficulty to the implementation of agroforestry systems in areas without traditional agroforestry use is to modify stocking rate at the level that avoids tree damage, as this component is profitable in the long term and animal numbers are not usually modified within the system. Adequate technical advice to farmers on how to manage the land appropriately is very important.

## Conclusions

Agroforestry systems probably have an important impact as a sustainable land use option, mainly as silvopasture and silvoarable systems.

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# Home-grown forage use on commercial dairy farms in the Basque Country

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## Abstract

Increasing the use of home-grown forage on dairy farms could contribute to improved feed self-sufficiency and reduced feed-N inputs from concentrates. 64 commercial dairy farms were sampled from September 2003 to April 2004 in order to describe lactating herd rations and to relate the home-grown forage use to land availability, milk production level, N inputs and N excretion. In this study, 26.6% of the sampled herds usually grazed and they were related to farms with lower livestock density or milk quota ( $P < 0.05$ ). Home-grown forage was basically composed of both grass silages and maize silages and their use reached on average 27.2% of ration dry matter. Grass silage was found in 90.6% of the 64 sampled rations representing on average 20.0% of the ration. Grass silage use was dependant on farm intensity ( $P < 0.05$ ) because silage use was higher when farm intensity was lower. Higher grass silage presence in the ration was related to higher grassland availability ( $P < 0.05$ ) and did not show a relation to milk yield ( $P > 0.05$ ). Silage quality should be reviewed in order to reduce concentrate inputs because of low RFV indexes (Relative Feed Value) observed. Low intensity farms (lower use of imported N) showed higher total N excretion per milk unit ( $P < 0.05$ ) but when N excretion per milk unit was referred to imported/purchased N excretion per milk unit there were no differences between intensification levels ( $P > 0.05$ ). Further, imported N excretion per herd and hectare also decreased with a lower intensification level ( $P < 0.05$ ).

Keywords: homegrown forages, dairy production, survey

## Introduction

Grassland has different degrees of predominance and contributes in different ways to milk production depending on the farming system used, which is often based on local traditions (Kristensen *et al.*, 2005). Basque traditional dairy farming activity has been historically carried out by non-specialized farms in which cattle were basically fed through grazing management from spring to autumn and grass hay-based rations in winter. Nevertheless, the farming model in the territory has changed towards more industrialized commercial farms. This strategy has involved the substitution of both grazing management and grass hay production by more usual grass silage use as main home-grown forage (INIA OT00-037-C17 project). This trend has been reported in other European countries where the role of grassland has also changed due to intensified livestock production and currently is playing a marginal role (Kristensen *et al.*, 2005). Further, intensified dairy farms tend to enhance nutrient inputs from concentrates which have been linked to increasing environmental pollution (Tamminga, 1992). The objectives of this study were to describe lactating herd rations in The Basque Country and to relate the home-grown forage use to land availability, milk production level and imported-N excretion by the herd.

## Materials and methods

A survey was conducted between September 2003 and April 2004. Four Milk Producers Advisory Centres selected 64 Holstein commercial dairy farms representing different ranges of milk yield (from 5,541 kg cow<sup>-1</sup> year<sup>-1</sup> to 12,166 kg cow<sup>-1</sup> year<sup>-1</sup>) and intensification levels based on yearly milk production per hectare (low < 12,000 kg ha<sup>-1</sup>; 12,000 kg ha<sup>-1</sup> < medium > 15,000 kg ha<sup>-1</sup>; high > 15,000 kg ha<sup>-1</sup> by Berentsen *et al.*, 2003). These farms also were selected due to their economical feasibility. Farmers were contacted by telephone and later interviewed face-to-face. The survey included information on adult dairy herd characteristics, milk production level and quality parameters (if known), milking herd rations, nutritional management strategies (grazing activity) and finally the land area and its use. On-site sample collection was carried out to complete a nitrogen balance at lactating cow level. On-farm sampling included feed, faecal, urinary and milk samples. Further, each herd was simulated with Cornell Net Carbohydrate and Protein System 5.0 model (CNCPS) in order to estimate faecal and urinary excretion volumes. All samples were analysed for N-Kjeldalh content. Daily N intake was estimated based on N concentrations of individual feeds and ration information. Freshly deposited faeces and urine (representing 5%-15% of the herd) were sampled in the barn. Daily faecal and urinary N excretion levels were estimated by multiplying estimated excretion volume and sample N content. After total N excretion determination, imported-N excretion was estimated applying the percentage of imported N intake according to Powell *et al.* (2002). Statistical analyses were carried out using Statview software (SAS Inst Cary, NC). Treatment means were tested using regression models for continuous variables or ANOVA for discrete variables after checking the normal distribution of data. Tukey-Kramer test was used as *post-hoc* analysis with significance level of  $P < 0.05$ .

## Results and discussion

Descriptive parameters of surveyed dairy farms are shown in Table 1. These data showed that on average sampled farms in this study are more intensified than those reported by Kristensen *et al.* (2005) in different European countries. Sampled farms did not represent the average dairy farm in The Basque Country, where mean milk production was 7401 kg year<sup>-1</sup> and herd size was 32 cows in 2003-2004 (Eustat, 2004). The four Milk Producers Advisory Centres in the region selected those farms biased to a more industrialized model in order to represent the next future production system of dairy farming in the territory.

Table 1. Descriptive parameters from 64 commercial dairy farms.

Parameters	Mean	Std
Cow herd size	87	76
Milk yield, kg cow <sup>-1</sup>	9,057	1,395
Farmland, ha	47.9	18.6
Grassland, ha	42.9	30.2
Cropland, ha	5.0	11.7
Density, cow ha <sup>-1</sup>	2.1	1.2
Milk yield, kg ha <sup>-1</sup>	16,767	9,522

One consequence of the intensification process is that grazing activity is reduced. Only 26.6% of sampled herds usually grazed from March to October and the grazing management was related to a lower livestock density or lower milk quota farms ( $P < 0.05$ ). In this sense, Parsons *et al.* (2004) found that herd size was the most significant factor between confinement and grazing farms. Home-grown forage was basically composed of both grass silages and maize silages and their use reached on average 27.2% (SD = 18.1%) of lactating herd ration. It is noteworthy the use of grass silages because 90.6% of surveyed farmers assured its use. The mean use of grass silages in the rations was 20%, slightly higher than maize silage (15%)

and alfalfa hay (15%) use and the mean forage:concentrate ratio was 50:50 for all the rations. Grass silage use was dependant on farm intensity because the higher grass silage presence in the ration was related to higher grassland availability ( $P < 0.05$ ) and smaller herd size ( $P < 0.05$ ). Milk yield did not show statistical relation to grass silage use ( $P > 0.05$ ). Bearing in mind the importance of grass silages it would be advisable to review their quality. Thus, RFV (Relative Feed Value) index was applied for all grass silages. Results showed that two out of three silages were allocated from Third to Fifth categories (RFV  $< 102$ ) and it was remarkable the high percentage of silages with the lowest quality index (20.5%). Furthermore, a univariate regression model showed that herds fed with high quality grass silages showed higher annual milk yields ( $P < 0.05$ ). On the other hand, diets consisting of grazed grass have been associated to low efficiency of protein utilization and therefore high N excretion (Hoekstra *et al.*, 2007). In this sense, data reported showed that farms which used mainly home-grown forages (low intensification), had a lower N-efficiency in milk and therefore total N excretion per produced milk unit was higher ( $P < 0.05$ ; Table 2). However, together with total N excretion it would be also advisable to consider imported or purchased-N excretion per milk unit because imported N means the constant nutrient input not coming from animal-soil-plant system. Purchased N was mainly imported in the form of concentrates and imported alfalfa hays. Most intensified farms imported more N than less intensified farms ( $P < 0.05$ ) and therefore when N excretion per milk unit was referred to purchased-N excretion, there were no differences between intensification levels ( $P > 0.05$ ) (Table 2). Furthermore, when yearly imported-N excretion data were considered together with herd and hectare variables, imported-N excretion decreased with lower intensification level ( $P < 0.05$ ) (Table 2).

Table 2. N excretion parameters for high (HI), medium (MI) and low (LI) intensified farms.

Parameters	Mean (sd)	HI	MI	LI
N <sub>total</sub> excretion/milk unit, g kg <sup>-1</sup>	14.9 (2.7)	13.8 <sup>a</sup> (2.4)	14.6 <sup>a</sup> (3.0)	16.4 <sup>b</sup> (2.4)
N <sub>imported</sub> excretion/ milk unit, g kg <sup>-1</sup>	11.7 (2.7)	12.1 <sup>a</sup> (2.5)	11.2 <sup>a</sup> (2.8)	11.4 <sup>a</sup> (3.0)
N <sub>imported</sub> excretion/cattle/land area, kg ha <sup>-1</sup> year <sup>-1</sup>	273.3 (165.0)	359.1 <sup>a</sup> (160.6)	181.9 <sup>b</sup> (62.4)	137.7 <sup>b</sup> (51.5)

Columns followed by different upper case are different at significance level  $P < 0.05$ .

## Conclusions

Grazing has decreased in the Basque Country in response to the intensification process that has occurred, as in most European dairy farms. Thus, grasslands are used for silage production and its use is limited by herd size and farmland area. Furthermore, as land base production is low, intensification is associated with lower home-grown forage N use and higher levels of purchased-N excretion per hectare. Taking into account the importance of grass silages in an intensified system, their quality should be reviewed and improved.

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# Citizens' evaluation of rangeland resources for a sustainable regional development

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## Abstract

Natural rangelands could be the source of many ecological, social and financial benefits and are of great importance to rural development. This paper presents an analysis of a citizens' evaluation on rangeland resources and their impact on regional development. The study was conducted in the prefecture of Boiotia, central Greece, through the use of a questionnaire. All collected data were subjected to statistical analysis. The majority of the public believed that grazing is the best way to manage rangelands. Some believed that rangeland resources should be managed for environmental purposes (enhanced biodiversity, erosion control, etc.) and for the development of ecotourism. It is also believed that the livestock products produced by grazing are of better quality than those produced by indoor feeding. Most of the Boiotia citizens believed that the state should consider public opinion on the management of rangelands.

Keywords: attitudes, rangelands, multifunctional

## Introduction

It has been proved that not only in remote and unfavourable regions, but also in the most homogeneous rural regions, the natural resources contribute considerably to their economic development (Schraml *et al.*, 2002). In the past, the accent was on the primary production of natural resources for the improvement of the rural economy. Nowadays, it is on the promotion and strengthening the ecological and social values that they offer (Elands and Wiersum, 2001).

Rangelands constitute the largest part of the earth's terrestrial natural resources. In the Mediterranean region they represent 52% of the area, and in Greece 40% of the total (NSSG, 1994). The multiple uses of rangeland ecosystems that contributed to the improvement of husbandry, wild life and environment is of great ecological and economic interest (Holechek *et al.*, 1989). Comprehending the perceptions of local communities and how they vary may be critical to managing rangelands. A better understanding of rural people's knowledge of rangelands and their impact on the socioeconomic and environmental welfare, is fundamental to the development and implementation of management strategies that are both sustainable in the long term and sensitive to existing local needs.

This paper presents an analysis of citizens' evaluation on rangeland resources and their impact on regional development.

## Materials and methods

The prefecture of Boiotia in central Greece was selected as the area of study. The population of the prefecture is 124,000 people (NSSG, 2003). Boiotia prefecture was selected as a study case as it is a traditionally pastoral region, and it borders with the prefecture of Attica where

Athens the capital city of Greece is. The prefecture covers 295,200 ha, 110,000 ha of which are plains.

Boiotia prefecture is highly developed agriculturally. Sheep and goat numbers are approximately at the average of Greece prefectures (Table 1).

Table 1. Livestock numbers in Boiotia prefecture in 2003 (NSSG, 2004).

Sheep	139,545
Goats	110,353
Total cattle	7,177

The research was performed through the use of a questionnaire. The method of face-to-face interviews was used, which is one of the best approaches of obtaining statistical data (Daoutopoulos, 2005).

The sampling method used was the simple random one due to its simplicity and because it requires the least possible knowledge regarding the population of any other method (Matis, 2001). As the variables refer to proportions, the determination of the total sample size is provided by the formula:

$$n = \frac{t^2 p(1-p)}{e^2}$$

where p = the estimation of proportion, p = 0.5

t = the value of the STUDENT distribution for probability (1-α) = 95% and n-1 degrees of freedom. In practice, for 95% probability the value is 1.96 (Matis, 2001).

e = the maximum admitted difference between the sampling mean and the unknown mean of the population (0.05). Based on the above formula, 385 valid questionnaires were collected. Data were subjected to analysis using the SPSS programme.

## Results and discussion

The majority of the questionnaire respondents (55.3%) consider that livestock grazing is the most suitable way for rangeland management (Table 2). A very high percentage (70.6%) of the respondents considers that the products of extensive livestock farming are of better quality than the ones of intensive farming. This view is probably influenced by the recent series of food scandals (Bovine Spongiform Encephalopathy, Poultry Flu, Dioxins) associated with the intensive livestock farming and to the increasing sophistication of consumer preferences compared with the past due to financial and social changes (Patterson *et al.*, 1995). Only 3.7% have the opposite opinion about extensive livestock-farming products.

It is particularly important that quite a few respondents acknowledge the multiple role of rangelands and the services and the goods that they offer concerning the environment, and for hunting and recreation. The contribution of rangelands to hunting, aesthetic value of landscape and recreation is recognized by 63.1%, 61.8% and 31.7% respectively, while 68% consider rangelands as lands of high historical and cultural value. Their contribution to protection from erosion is recognized by 84.9%.

Almost all the rangelands in Greece are public. Therefore it is not surprising that the majority of the respondents (54%) wish their opinion on issues concerning the management of rangelands to be taken into consideration. The participation of citizens in the management of rangelands is critical in terms of compatibility with local development. In many cases the participation is only typical, and critical decisions are taken by closed bureaucratic bodies and/or by central organizations. Thus, the active participation of the local population in the

decision-making on rangeland management, the participation in the management itself and in the whole range of benefits that derive from it, becomes essential.

Table 2. Evaluation of rangeland resources by citizens (%).

	Strongly disagree	Disagree	Neither agree nor disagree	Agree	Strongly agree
1. Livestock grazing is the most suitable way of rangeland management.	5.7	7.5	31.4	30.6	24.7
2. The products of extensive livestock-farming are better quality than those of intensive farming.	0.8	2.9	25.7	36.1	34.5
3. Significantly contribute to hunting.	10.4	5.2	21.3	38.7	24.4
4. Increase the beauty of the landscape.	3.6	13.0	21.6	27.8	34.0
5. Provide few opportunities for recreation and exercise.	12.5	28.6	27.3	20.5	11.2
6. Have important cultural and historical value.	3.9	6.8	21.3	32.2	35.8
7. High protective importance (for floods etc).	1.0	2.9	11.2	30.9	54.0
8. Citizen's opinion on issues concerning the management of rangelands should be taken into consideration.	19.5	11.7	14.8	31.9	22.1

## Conclusions

This study provides evidence that people grant considerable importance to the economic and environmental benefits of rangeland resources. Furthermore, the participation of citizens in the management of rangelands is critical in terms of compatibility with the local development.

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# Efficient utilization of grassland biomass for energy purposes through mechanical dehydration of silages

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## Abstract

The cultivation of grassland especially in low mountain ranges is facing great difficulties. The main reason is the reduction of animal husbandry. However, the botanical diversity and other ecological functions of grassland can only be maintained with a regular defoliation and care. Hence, new concepts are required, and the energetic use of grassland biomass may be a new source of income for farmers. Yet, in common biogas plants, conversion from grassland biomass into biogas is difficult because of the high lignin content, and the combustion of hay is exceedingly difficult due to the increased ash contents and the resulting emissions. The mechanical dehydration of ensiled biomass prior to the anaerobic digestion provides a promising alternative. The quality of the biomass is less significant. One important aim of this process is the reduction of minerals in the presscake, which will be used as solid fuel. The K-content of extensive grassland biomass was reduced with mechanical dehydration at more than 50%. The combustion of the biogas covers the heat demand for drying the solid phase to a storable fuel. Compared with the original biomass, the quality of the pressed solid fuel is significantly improved due to reduced contents of minerals.

Keywords: grassland, low mountain range, energy use, solid biofuels

## Introduction

Climate change and the higher costs and the shortage of fossil fuels are leading to a strong increase of the energetic use of biomass. This competes with the utilization of biomass for fodder or foodstuff. On the other hand, the cultivation of grassland in low mountain ranges is at risk due to the decline in animal husbandry. Those grasslands are often very rich in species, which are very important also for faunal biodiversity. But the diversity of grassland can only be maintained through a regular defoliation. The energetic use of grassland biomass could be a quite interesting perspective, as it would not compete with other crops. For grassland biomass two conversion routes are discussed: anaerobic fermentation (biogas production) and thermal conversion (combustion of hay). The combustion of hay with its high content of minerals is difficult, particularly due to emissions and ash problems. The anaerobic fermentation of grassland biomass in common biogas plants is difficult due to high lignin contents, which increases the retention time in the reactor. It is the biomass of extensive grasslands in particular that often has higher lignin contents, as cutting dates are delayed in order to allow herbs to flower.

Hence, new concepts are required, and biogas production from grassland with an innovative technique may be a perspective. The mechanical dehydration of ensiled biomass prior to the anaerobic digestion provides a promising alternative, as it is quite flexible regarding the quality of the biomass. The technique, which has been developed at the University of Kassel (Reulein, 2007; Wachendorf *et al.*, 2007) is displayed in Figure 1. After the mechanical dehydration of the silage the press cake is available for thermal conversion. The press fluids serve as substrate for biogas production with the final products electricity and heat.

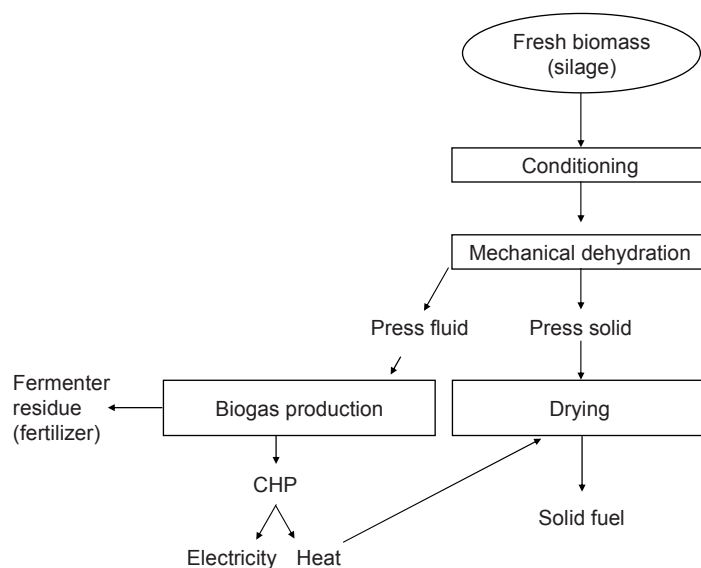


Figure 1. Principle of the integrated biogas and solid fuel production from agricultural bio-masses.

For direct combustion, the biomass from species-rich grassland (preferably as hay) is not suitable due to its high contents of minerals. By mechanical dehydration the biomass is significantly upgraded. In addition to a reduced water content the lower ash contents represents a substantial improvement, especially for use as solid fuel, due to the prevention of corrosion damage evoked by chloride and potassium. The reduction of nitrogen in the press solid leads to a lower discharge of nitrogen oxides during combustion. Drying of the press cake to a storable and transportable level is attained by using waste heat resulting from electrification of biogas resulting from the press fluids. The biogas production with press fluid, which mainly contains easily fermentable components, leads to high methane yields between 450 and 500 litre CH<sub>4</sub> per kg oDM. Organic matter in the fluids from maize and grass silage was fermented by 90%. The fermentation of the press liquids takes a maximum of 4 days (Reulein *et al.*, 2007). Studies on this conditioning process have been performed with the objective of describing mass flows of silage constituents into the press cake and press fluid (Reulein, 2002). The results suggest that by dehydration with a worm extruder, 25 to 30% of the DM contained in the whole silage (maize) is transferred into the press fluids. Thus, one-third of the chemically bounded energy in the DM of the silage is transferred into the press fluids and two-thirds into the press cake. Furthermore, the easily fermentable components of the silage (sugars, proteins, etc.) go in the press fluid, whereas lignin and other low-fermentable components remain in the press solid. After pressing, the OM content of the press solid is approximately 50%. Of the crude ash (XA) in the silage, 60-70% is transferred into the press fluids. The mass flow of nitrogen (N) into the press fluid accounts for 35-50%. In the present paper, data on mass fluxes (OM, N, P, K) into the liquid and solid phase and on the quality of the solid fuel produced are presented from experiments with grassland biomass from low mountain range.

## Materials and methods

Ensiled biomass from extensive, semi-natural grasslands in low mountain range (Black Forest in southern Germany and Rhön in the middle of Germany) served as substrate for the experiments. They comprised mostly lignified forages from typical grassland communities (e.g. *Arrhenatheretum*, *Trisetetum*, *Nardetum*, different tall-herb communities, etc.) harvested



at the end of July. The ensiled biomass from 3 replications was mixed and mashed under different temperature levels between 5 °C and 80 °C with water at the rate of 1:5 without any replications. After the mashing, the biomass was mechanically drained with a worm extruder. DM-, N-, P- and K- contents were determined in the raw material, in the press solid and in the press liquid. The amount of press solid and press liquid was calculated (based on DM) in relation to the raw material and mass flows of the different constituents (oDM, N, P, K) in the press liquid were determined.

## Results and discussion

N, P, K and oDM fractions found in the press liquid (Figure 2) were obviously affected by the conditioning of the silages. The diagram gives average values across all grassland types. An increased mass flow into the press liquid could be achieved for all examined constituents by a 15-minute mashing at increased temperatures, compared with mashing with cold water (5 °C). Mashing with water of 60 °C lead to a mass flow into the press liquid of 84% of K, 72% P, 38% N and 30% DM. In the case of N, an increase of the mash temperature up to 80 °C achieved a further (but small) increase in mass flow.

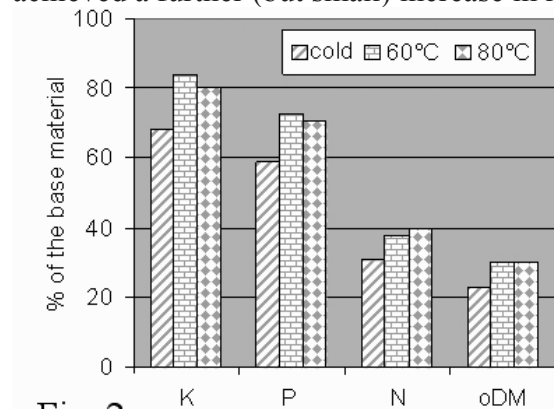


Fig. 2

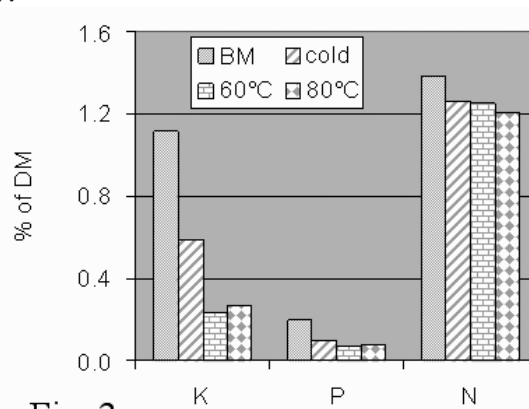


Fig. 3

Figure 2. Mass flows of N, P, K and oDM into the press liquid of extensive grassland (% of base material) as affected by mashing with different temperatures prior to mechanical dehydration

Figure 3. N, P and K content in the base material (= original material) (BM) and in the press solid of extensive grassland as affected by mashing of the ensiled biomass prior to mechanical dehydration

Figure 3 compares N, P and K content of original materials (BM) and press solids after different conditioning procedures. It appears that a short cold mashing at 5 °C induced a reduction in the press-solid mainly for K, which could be further enhanced by a 15-minute mashing at 60 °C. Mashing at 80 °C caused no further reductions.

The integrated biogas and solid fuel production provides interesting aspects for an economical and ecological reasonable utilization of grasslands in low mountain ranges and other rural areas. Compared to original biomass, the chemistry of the solid fuel can be significantly improved by mashing the silage followed by the dehydration with a screw press.

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# Impact of global climate change scenarios on alfalfa production in France

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## Abstract

The impact of global climate change is a problem for stockbreeders: if forage production is low for many days or weeks, it is necessary to provide hay or other stored feed. The aim of this work is to estimate the magnitude of changes in level and dates of production of an alfalfa crop. Growth simulations were made using the crop model STICS, and two distant IPCC scenarios for two future periods. We first studied the climate change alone and then both the climate and CO<sub>2</sub> effects, bearing in mind that the assumptions on model reliability are different. We created weather series using the method of anomalies and we present here mean results for one location. The main result is the positive balance of the effects of CO<sub>2</sub> and warming associated with droughts.

Keywords: climate change, alfalfa, grass, production

## Introduction

The impact of global climatic change is a source of concern for stockbreeders because the management of food supply for livestock depends on the seasonal or daily grass or legume production. If the production is low for many days or weeks it becomes necessary to provide hay or other conserved feed. The aim of this work is to estimate the importance of changes in level and dates of production. We will use the example of one crop, alfalfa, chosen because it is now an expanding crop in France according to the records of the network of some 1,500 breeding farms managed by the Institut de l'Elevage, mostly in the south of Europe. Another advantage of alfalfa is that production from this crop will not be limited by lack of nitrogen as a result of an increased demand for fertilizer nitrogen if production increases. Under climate change scenarios the climate will be warmer and drier; it therefore seems highly probable that the summer production will decrease. The main question is about the possible increase of the autumn production, with later cuts, involving changes in the herd management or feed supply.

## Materials and methods

As climate data, we use the *ARPEGE climat* scenarios (from Météo-France) also used in the IPCC studies, and results from just two IPCC scenarios, A2 and B1, for the near (2020-2049) and distant future (2070-2099), chosen for a moderate (B1) and high (A2) CO<sub>2</sub> increase. Weather records were created using a local reference series (1980-2006) and differences between present and future simulated series (Huard, 2007), giving a 27-year series.

Growth simulations were made using the crop model STICS (Brisson *et al.*, 1998, Ruget *et al.*, 2006) studying firstly the climate change only, and secondly climate and CO<sub>2</sub> effects. This method makes use of our knowledge of the effect of each component. The model takes into account climate, and we assume that all the functions and parameters remain valid in the future climatic context. The functions dealing with the effect of CO<sub>2</sub> are the effect on photosynthesis (which increases as a function of CO<sub>2</sub> concentration) and the effect on transpiration, which decreases because of the increase in stomatal resistance due to the high CO<sub>2</sub> concentration. The plant features were estimated using 10 year x location situations in France, with 3 or 4 regrowths for each, and more or less available water for the crop (without



irrigation in a dry summer climate). One advantage of a legume crop (in this case alfalfa) is its symbiotic nitrogen fixation, so that there is no (or only a small) nitrogen deficit in the current climate. The cutting date is determined by the heat sum (base 0°C) after February 1<sup>st</sup>, in order to take advantage of the global warming, and a minimum harvestable dry matter yield of 0.5 t ha<sup>-1</sup>. The model does not include any function for summer mortality of plants. The simulations were made for 6 locations and 2 soil types (available water AW 66 and 208 mm) at each location, 3 forecast numbers of cuts, i.e. 3 sets of intervals between cuts.

## Results and discussion

We refer here to only one meteorological station, at Carcassonne (02°22'E, 43°12'N), in south-western France, a warm but reasonably well-watered region at present, with mainly shallow soils. The climate, represented by mean monthly values, will be hotter, especially in the distant future and the A2 (high CO<sub>2</sub>) scenario, as the highest mean monthly temperature over 27 years will reach 29 °C. The precipitation will be higher in winter (except in and around the B1 scenario) and lower in summer in the future, except in the near future when the mean forecast rainfall will be higher for almost every month. The basic water balance (P-ETP) will be negative for the March-October period, but the deficit will be larger in the distant future.

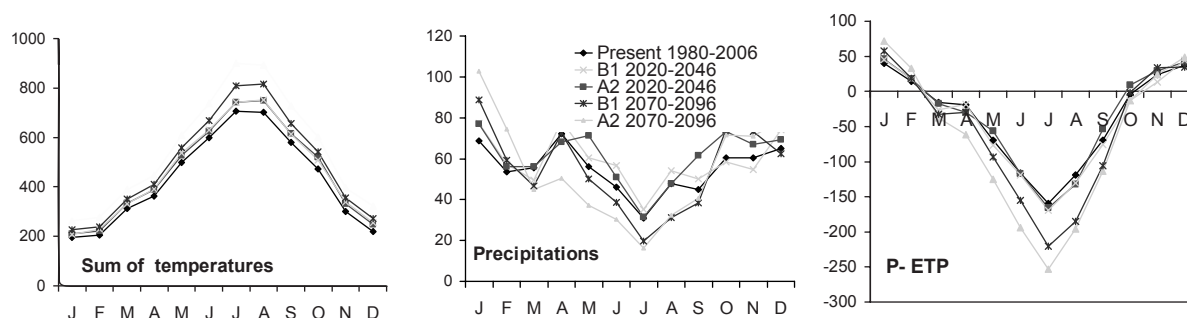


Figure 1. Monthly mean sums of temperatures (> 0°C), precipitation and water balance (P-ETP for present and future climate series (B1 and A2) each for the near and distant future.

Concerning the agronomic variables, the dates of the first cuts will become earlier, because of the temperature increase that accelerates leaf development, giving the same production in a shorter time. There is no decrease in the spring cut because there is no water stress during this period. The dates of the second and third cuts become earlier in the near future and generally later in the distant future. In the distant future, the last cut is even later because of slow growth due to lack of water at the end of the season. That means that the timing of the last cuts depends not only on available temperature, but mainly on available water. The frequency of the last cut is never 100%, i.e. it is not possible to make four or five cuts either now or in the future. The fourth and fifth cuts will be delayed and the proportion of years with late cuts reduced from now to the near or distant future.

Table 1. Production of alfalfa for 5 scenarios, with and without the effect of CO<sub>2</sub>.

Mean annual production (t ha <sup>-1</sup> )	Present	B1n	B1d	A2n	A2d
CC	7.54	7.84	7.86	7.21	6.72
CC+CO <sub>2</sub>	7.54	8.43	8.68	8.37	9.18

As a consequence of the climatic effect, the production of each cut will decrease. Depending on the scenario, production increases (B1) or decreases (A2). The extra CO<sub>2</sub> increases production, especially in the 'distant A2' combination, where CO<sub>2</sub> concentration is high. In the near future, without the effect of CO<sub>2</sub>, production is slightly altered, depending on the

scenario, whereas it is reduced in the A2 distant future by less than 10%. The small increase in production is caused by the increase in temperature, without any decrease in water availability (due to an increase in precipitation). Even without mortality simulation, the future decrease in summer (A2) cannot be completely compensated for by the autumn or winter production. With the effect of CO<sub>2</sub>, production is always slightly increased (10-20%).

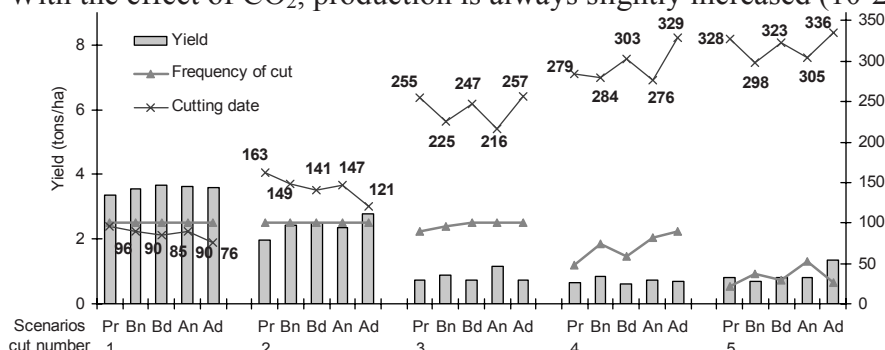


Figure 2. Yield (tonnes DM ha<sup>-1</sup>), cutting dates (day number from Jan. 1) and frequency (maximum number of years = 27) of each cut in Carcassonne, for 5 scenarios (present, B1 and A2 in the near (n) and distant (d) future, taking into account climate change + direct CO<sub>2</sub> effects). Yields and cutting dates are mean values for 27 years.

Therefore, this effect depends largely on parameters for stomatal behaviour, which are not very well known for field conditions. Hence these results can only be regarded as probable trends for future production. In all the cases, the deep soil gives higher production (about double) than the shallow soil, showing that productivity is reduced by the lack of water in the shallow soil. The effect of climate change, even with the effect of CO<sub>2</sub>, is much less than the effect of soil depth and water reserve. One main result is the advancement of production and harvesting dates, associated with a longer period without production in summer. As compensation for the low summer production, it seems to be rarely possible to take an additional cut, except on the deep soil. This means that the lack of production is due to water shortage. The reduction of water availability is more important than the temperature changes. For mown crops (grass or alfalfa), results are generally similar to these (Lüscher *et al.*, 1998; Soussana and Lüscher, 2007), as opposed to annual crops, which will generally have a lower production due to a shorter production period in the warmer future.

## Conclusions

The main results concern the dates of feed availability for livestock, which will be earlier in the future. In spite of the value of the results, it is absolutely necessary, when using the model, to be wary of the results, because they depend greatly on the reliability of the ecophysiological laws and most of all on the parameter values, which are not tested for the future conditions.

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# Perspectives of precision agriculture on grassland

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## Abstract

Precision agriculture has become well established in the past decades on arable land, whereas on grassland its introduction still faces considerable problems. One major constraint is the lack of sensor technology that precludes the rapid detection of sward characteristics that the management should react on. For example, sensor-based yield determination on forage harvesters can be well developed in pure stands of forage grasses, but fail to provide reproducible data in heterogeneous permanent grassland. However, some developments are promising and currently tested within the EU and abroad for possible applications, such as yield estimates and identification of vegetation types based on remote sensing. The paper focuses on current research issues and demonstrates the need for transdisciplinary work with unrelated research teams to promote precision agriculture on grassland.

Keywords: precision grassland agriculture (PGA), perspectives, constraints

## Introduction

Precision agriculture (PA) can be defined as the use of information technology in all of agriculture (Plant, 2001). This includes not only site-specific management of agricultural fields but also every agricultural activity that implements up-to-date information technologies along with plant production, animal production and welfare (precision livestock agriculture = PLA), agricultural landscape management and post-harvest processing of raw material (Stafford, 2000). PA is innovative, integrated and for the most part internationally standardized (Cox, 2002). PA is technology driven and so it developed rapidly throughout the past two decades when personal computers and chip technology enabled the ease of handling huge data sets.

## Managing heterogeneity

PA technology has mainly been implemented in arable cropping whereas its introduction on grassland still faces considerable problems. The heterogeneity of grassland fields in terms of floristic composition, sward characteristics, standing dry biomass, and forage biomass is the major constraint to the application of existing PA routines. Our inability to explain such heterogeneity sufficiently hinders the introduction of techniques allowing to react adequately on it. Heterogeneity is not only timely but also spatially displayed. Frequent utilization of grassland sward changes that spatial heterogeneity as well as fertilizer application does. It has been shown that spatio-temporal heterogeneity changes considerably on permanent grassland (Bailey *et al.*, 2001). To consider that change when applying technical applications remains the future challenge of research on precision agriculture on grassland.

## Image processing

Evaluating current developments of PA on arable land helps to consider their possible application on grassland. In many applications the recognition of objects (mainly plants and sward properties on grassland) is a key issue that has led to an increase in experimental work on sensors and on digital image analysis (DIA). On grassland, only few has been published on

the detection of individual species (Bonesmo, 2004; Gebhardt *et al.*, 2006; Schut, 2006). The problem is that the soil background to which the plant contour and colour could be discriminated is not visible in most cases. Moreover, small scale changes in colour and shape of the objects occur in dense grassland swards with multiple overlapping. One possible approach is that of a segmentation of objects from images based on colour and grey level intensity in the visible and near infrared range of the light spectrum. Shape, texture and colour parameters can then be used to classify grassland species (Gebhardt *et al.*, 2006). For *Rumex obtusifolius* that approach worked well and hence potentially allows site-specific spraying of herbicides to areas where this species has been detected. A future perspective of DIA application of grassland can be that of an automatic detection of species on-the-go and a parallel automated spraying system linked to DIA.

### **Remote sensing**

Today's technique allows the gathering of valuable ground information on grassland properties from a remote position above grassland fields. On arable land, remote sensing has already become well established where a wide range of physical, chemical and area based parameters can be detected. Progress has also been made in the recognition of grassland types (Schmidtlein *et al.*, 2007) and nutritional status. However, applications on grassland are still rare. The question remains still open as to whether remote sensing can contribute to the identification of small scale heterogeneity that can be managed by precision agriculture technology. Spatial resolution, return rate of the sensor (mostly satellite borne), and cloud cover are most limiting.

### **Yield measurements and site-specific management**

The automated measurement of grassland yield at given locations is a prerequisite of managing grassland site-specifically. But, measuring grassland yield without laborious and time consuming clipping technique is difficult to put into practice. Measurements of flow rate and water content have successfully been performed only in pure stands of ryegrass with near infrared spectroscopy, and there is no applicable technique available to roughly estimate local yield except the rapid pasture meter ([www.farmworkspfs.co.nz](http://www.farmworkspfs.co.nz)). Considerable research demand has been identified when screening the scientific literature on automated yield measurement systems. On the other hand, such techniques would help to fill an important gap in one's knowledge with respect to fertilizer application, feed management, grazing practice, and environmental protection.

Most applications of PA focus on managing zones within an agricultural field that requires variable adjustment of crop treatment, i.e. site-specific management. Each management zone can be expressed as a homogenous combination of yield determining factors for which a single rate of a specific crop input or management intervention is appropriate (Doerge, 1998). On grassland, it is challenging to define such zones because sensors must be developed to replace laborious and time-consuming sampling and measurements on plants and to enable automatic sensing of canopy and sward properties. Once ready for practical use, site-specific management on grassland would mainly be applied to spread fertilizers and, to a much lower extent, to spray herbicides. However, the ecological and economic benefit of such management practise still remains unclear.

### **Challenges**

Precision agriculture has developed to an autonomous multidisciplinary area of expertise which is recognized by the scientific community through journals and numerous congresses all over the world. However, on grassland precision agriculture has not yet found its place. It

is obvious that cooperation of grassland scientists with technical engineering is necessary to bring precision agriculture forward. By doing so, grassland scientists can bring in their expertise about impact of habitat, environment and management on grassland characteristics and productivity.

Application of precision agriculture on grassland still faces numerous technical problems mainly in the field of sensor technology to detect sward properties. So, transdisciplinarity is required, in other words: grassland scientists have to involve several unrelated academic disciplines to reach a common research goal (Tress *et al.*, 2007). Beforehand, scientific goals need to be defined and chances for conversion into farm practice must be assessed.

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# Suitability of low-intensity grassland for combustion as influenced by grassland community and harvest date

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## Abstract

Since semi-natural, biodiversity-rich grasslands increasingly cease to play a role in animal nutrition, alternative uses for the biomass have to be found. The co-combustion in existing straw-firing systems is a promising option. However, the combustion of grassland biomass poses considerable challenges due to its chemical composition. The chemical parameters relevant to the combustion process may be expected to improve with increasing plant maturity and also to differ between grasslands of different botanical composition. Therefore, biomass from five semi-natural grassland communities of different moisture regimes and nutritional status were harvested at four dates from June to September 2007 and the development of their chemical composition was studied. Grasses and forbs were analysed separately for N, S, Cl, K, Ca, and ash content. Grasses had lower N, Ca, and ash, and higher Cl contents than forbs. Between June and September N, Cl, and K contents decreased, while Ca and ash contents increased, leading to a slight overall fuel quality improvement. Site effects were particularly marked for K, Cl, and S. While an entirely satisfactory fuel quality could not be attained by the investigated biomass fractions, biomass from most sites could probably still be used for combustion in appropriate firing systems.

Keywords: grassland conservation, bioenergy, combustion, chemical composition

## Introduction

A large proportion of species-rich semi-natural grasslands in Germany is no longer needed for forage production. Its conservation is mainly achieved by state subsidies. Presently the harvested biomass has often to be disposed of. The production of bioenergy from hay in firing systems optimized for cereal straw would offer a utilization perspective for the future. A major obstacle for the use of hay as a biofuel is its chemical composition. Table 1 presents guiding values for relevant elements in biofuels. If these are exceeded the consequences include excessive emissions of NO<sub>x</sub>, SO<sub>2</sub>, and HCl (high N, S, and Cl contents), furnace corrosion (high K, Cl, and S contents) and slagging (high K and low Ca contents). As conservation grassland includes sites of different botanical composition, site characteristics and cutting dates, its chemical composition is also highly variable with the possibility of very problematic values for some of the above-mentioned parameters. As a consequence its use as a biofuel is difficult. The aim of the present study was to clarify the relative importance of botanical composition, site and cutting date on the suitability of hay from species-rich meadows for combustion.

## Materials and methods

The experiment was established as a randomized block design, with four harvest times as the experimental factor and four replications, at five sites of semi-natural grassland in south-west Germany (Table 2). Sites 1 and 2 represent dry calcareous grassland, sites 3 to 5 mesotrophic grassland of different moisture regimes (dry, intermediate and wet, respectively). At sites 1 and 2, the usual date for haymaking is in September, while at sites 3 to 5 a first cut is usually

taken between June and July, and a second between August and September. All sites had not been fertilized for several years except site 4, which occasionally received cattle manure. In the second decade of each month from June to September 2007, biomass was harvested by hand on four 1 x 1m-plots at each site. It was separated into grasses (*Poaceae* and *Cyperaceae*) and forbs (remaining species), dried, and ground to pass a 1 mm-sieve. Both fractions were analysed separately for N, S, Cl, K, Ca, and ash content. N and S were determined by combustion in a CNS analyser, K and Ca by flame photometry after microwave digestion, and Cl by ion chromatography after hot water extraction. Ash content was determined at 550°C. All analyses followed the methods of the VDLUFA (2002). The data were subjected to an analysis of variance. A model was fitted using the procedure GLM of the SAS/STAT 9.1 software, which included fraction (grasses/forbs), site and harvest date as the main factors as well as their interactions. Proportion of variance explained was calculated for all effects. Means were compared using Tukey's HSD.

Table 1. Threshold values for the dry matter content of selected elements in biofuels (adapted from Obernberger (1998)).

N	S	Cl	K	Ca
< 6 mg g <sup>-1</sup>	< 1 mg g <sup>-1</sup>	< 1 mg g <sup>-1</sup>	< 7% of ash content	15-30% of ash content

Table 2. Characterization of the 5 experimental sites including average DM yield over all 4 harvest dates.

Site	Vegetation type	Dominant species	Dry matter yield (Mg ha <sup>-1</sup> )
1	dry calcareous grassland	<i>Bromus erectus</i> , <i>Ononis spinosa</i> , <i>Lotus corniculatus</i>	2.04
2	dry calcareous grassland	<i>Bromus erectus</i> , <i>Ononis spinosa</i> , <i>Euphorbia verrucosa</i>	2.69
3	hay meadow, dry	<i>Arrhenatherum elatius</i> , <i>Bromus erectus</i> , <i>Rhinanthus alectorolophus</i>	2.10
4	hay meadow, intermediate	<i>Dactylis glomerata</i> , <i>Arrhenatherum elatius</i> , <i>Trisetum flavescens</i> , <i>Galium mollugo</i>	4.79
5	hay meadow, wet	<i>Holcus lanatus</i> , <i>Cynosurus cristatus</i> , <i>Juncus effusus</i> , <i>Ranunculus acris</i> , <i>Bistorta officinalis</i>	4.14

## Results and discussion

Average dry matter yields over all four harvest dates are listed in Table 2 for all sites. There was no significant change over time. In Table 3, the average chemical composition of grasses and forbs over all sites is presented for each harvest date. Table 4 shows the proportion of variance that was explained by each significant main effect or interaction in the ANOVA. Variation of N content was predominantly due to biomass fraction, with the N content of forbs being on average 58% higher than that of grasses. Within fractions, site differences were not consistent over harvest dates. A significant decrease occurred only for forbs. The threshold level given in Table 1 was in all cases exceeded. S contents showed a considerable variability, of which only a comparatively small proportion was attributable to the influence of factors under consideration. Those of forbs were marginally lower than those of grasses. The threshold value was exceeded for most individual samples. On average Cl contents were 44% higher in grasses than forbs. The highest average Cl content (6.83 mg g<sup>-1</sup>) was found on site 4, making biomass from this site clearly unsuitable for combustion. At the other sites, the threshold value could only be attained by few samples. K contents decreased by 26% on average between first and last harvest. For both grasses and forbs, they were highest on sites 2 and 4, while they were lowest on site 5. While on sites 1 to 4, K contents of forbs were 19-60% higher than in grasses, they were 38% lower than those of grasses on site 5. This may be attributed to the limited K supply at this site (Whitehead, 2000), which is indicated by the very low levels of herbage K in both fractions. The threshold level for K in biomass ash was only approached by the forbs fraction from site 5, while it was exceeded many times in the



remaining samples. Ca content in forbs was on average 3.7 times higher than that in grasses. In both fractions, there was a significant increase of 43 and 34%, respectively, between first and last harvest. Both for grasses and for forbs, the highest Ca content was found for the sites 1, 2 and 4, all having lime-rich soils. While Ca contents lay between 14 and 28% of biomass ash for all forbs, that value only ranged between 4 and 10% for grasses. Ash contents were higher for forbs than for grasses and increased significantly between first and last harvest for both fractions, although more markedly for grasses than for forbs.

Table 3. Chemical composition (mg g<sup>-1</sup>) of grasses and forbs at four harvest dates, average over all sites. Significant differences ( $P = 0.05$ ) within each fraction and each harvest date are marked by different lower case and capital letters, respectively.

		N	S	Cl	K	Ca	Ash
Grasses	June	13.96 aB	1.58 aA	3.98 bA	16.39 aB	4.50 cB	68.95 cB
	July	12.88 aB	1.61 aA	5.65 aA	15.18 aA	5.19 bB	77.84 bB
	Aug.	13.59 aB	1.64 aA	3.66 bA	13.50 bB	6.24 aB	80.11 abB
	Sept.	13.55 aB	1.65 aA	3.39 cA	12.04 cB	6.42 aB	82.97 aB
	$\bar{x}$ Grasses	13.49	1.62	4.17	14.28	5.59	77.47
Forbs	June	21.50 aA	1.41 aA	2.53 b	20.44 aA	17.36 cA	91.40 bA
	July	18.09 bA	1.53 aA	4.18 a	17.74 bA	18.85 bA	91.49 bA
	Aug.	17.74 bA	1.52 aA	2.43 b	16.02 bcA	22.18 aA	95.08 bA
	Sept.	16.51 cA	1.65 aA	2.46 b	15.11 cA	23.34 aA	99.67 aA
	$\bar{x}$ Forbs	21.50	1.52	2.90	17.33	20.43	94.41
Range		9.93-29.10	1.00-3.17	0.42-13.26	5.57-30.69	3.63-29.67	54.67-123.40

Table 4. Proportion of variance explained for main effects and interactions. Asterisks denote the level of significance.

	N	S	Cl	K	Ca	ash
Fraction	50.4 ***	2.4 *	6.5 ***	2.4 ***	91.8 ***	34.0 ***
Time	8.8 ***	ns	10.5 ***	9.0 ***	3.8 ***	7.9 ***
Site	4.5 ***	29.2 ***	49.3 ***	61.6 ***	2.6 ***	32.1 ***
Fraction x time	5.8 ***	ns	ns	ns	ns	1.3 ***
Fraction x site	8.9 ***	ns	0.6 ***	19.0 ***	0.2 ***	12.9 ***
Time x site	8.9 ***	12.8 *	26.6 ***	2.6 ***	0.2 *	1.7 *
Fraction x time x site	2.0 *	ns	0.9 **	ns	ns	ns

## Conclusions

On the sites investigated in this experiment, a high percentage of forbs would improve fuel quality with respect to Ca, Cl, and S content, while deteriorating it in respect of N, ash, and in most cases, K content. A perceptible improvement by a delayed harvest would be possible only for K and Ca content. K, Cl, and to a lesser extent, S and ash contents, were strongly influenced by site effects. Biomass from lime-rich soils with a low level of plant available K and no past fertilization with chloride-containing mineral fertilizer can be expected to have the most suitable fuel properties. Desirable contents of N, S, Cl, and K, listed in Table 1, could not be attained by any sample. However, given an appropriate firing system, biomass from most sites might still be used for combustion. As N, Cl, and ash contents, possibly most limiting for biofuel use, did not decrease markedly between June and September, harvest date could be chosen according to conservation requirements, creating real synergy effects between nature conservation and bioenergy production.

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## **Session 4B**

# **Forage-based animal production**

# Effect of pre-grazing herbage mass on production performance of dairy cows in mid-lactation

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## Abstract

The objective of this experiment was to investigate the effect of a high (H – 2100 kg DM ha<sup>-1</sup>) and low (L – 1500 kg DM ha<sup>-1</sup>) pre-grazing herbage mass (HM) on mid-lactation milk production from spring-calving dairy cows offered two levels of daily herbage allowance (> 4 cm) (16 kg or 20 kg DM cow<sup>-1</sup> day<sup>-1</sup>). Sixty-eight spring calving Holstein Friesian dairy cows were assigned to a 4 treatment (n = 17) grazing study (H16, H20, L16 or L20) from 4 April until end July. No supplement was offered. Pre- and post-grazing sward height and milk yield were recorded daily. Milk fat, protein, lactose content, liveweight and body condition score were recorded weekly. Herbage mass had no effect on any milk production parameters. Allocating extra herbage increased milk production performance by 0.25 kg milk per kg grass offered (23.1 kg (16 kg) and 24.1 kg (20 kg), respectively). Yield of milk solids ( $P > 0.01$ ) was higher with the high herbage allowance treatments. Higher grass utilization was achieved on the low herbage mass treatments. Preliminary results indicate that the balance of attaining high sward utilisation and high cow performance from pasture is achieved by using lower pre-grazing HM and allocating cows a high DHA.

Keywords: dairy cow, milk production, herbage mass, allowance

## Introduction

With future feed costs predicted to rise, a transformation in grassland management has taken place over the last decade. Increasing the proportion of grass in the diet of the dairy cows is now a major objective in grazing dairy systems. A number of sward factors have been shown to influence the rate of herbage intake of grazing animals. Stakelum (1986) reported increased intake as herbage mass increased. Hendricksen and Minson (1980) stated that the rate of intake is more closely related to green leaf mass than to sward height. As herbage mass increases, the proportion of green leaf in the sward decreases. The objective of this experiment was to investigate the effect of a high (H – 2100 kg DM ha<sup>-1</sup>) and low (L – 1500 kg DM ha<sup>-1</sup>) pre-grazing herbage mass (HM) on mid-lactation milk production of spring calving dairy cows offered two levels of daily herbage allowance (> 4cm; DHA) (16kg or 20 kg DM cow<sup>-1</sup> day<sup>-1</sup>).

## Materials and methods

The experiment was conducted at Moorepark Dairy Production Research Centre, Fermoy, Co. Cork, Ireland. The underlying soil type is a free draining acid brown earth of sandy loam-to-loam texture. The area used was under permanent pasture with a predominately ryegrass sward (*Lolium perenne* L.). The swards were on average five years old.

The experiment was a randomized block design with a 2 × 2 factorial arrangement of treatments. Sixty-eight (20 primiparous and 48 multiparous) spring calving Holstein Friesian dairy cows (mean calving date 10 February; s.d. 15.8 days) were randomly assigned to a

4-treatment ( $n = 17$ ) grazing study. The treatments imposed were two levels of pre-grazing herbage mass (HM) and two levels of daily herbage allowance (DHA). The treatments were high (H – 2100 kg DM ha<sup>-1</sup>) or low (L – 1500 kg DM ha<sup>-1</sup>) pre-grazing HM and high (20 – 20 kg DM cow<sup>-1</sup> day<sup>-1</sup>) or low DHA (16 – 16 kg DM cow<sup>-1</sup> day<sup>-1</sup>). The four treatments were H20, H16, L20 and L16. Treatments were imposed from 4<sup>th</sup> April until the end of July. From calving until treatment assignment, animals were grazed as one herd. No concentrate was offered during the experiment. The experimental area consisted of 20 paddocks which were randomized, and five paddocks were assigned to each individual treatment. All paddocks were grazed once prior to the beginning of the experiment in order to create differences in the pre-grazing HM. Fresh herbage (> 4cm) was allocated daily to each treatment following morning milking. Herbage mass and sward density were measured twice weekly in each grazing area by cutting four strips per grazing area. Pre- and post-grazing sward heights were measured daily. Milk yield was recorded daily for each individual animal. Milk fat, protein and lactose concentrations were calculated weekly. Bodyweight and body condition score (BCS) were also measured weekly. Data was analysed using covariate analysis in SAS (2002). The factors in the model were pre-grazing herbage mass (HM), DHA, interaction between HM and DHA and the appropriate pre-experimental variable.

## Results

Milk production results are shown in Table 1. There was no interaction between pre-grazing HM and DHA throughout the experimental period. There was no difference in milk yield between animals grazing the low HM swards (23.5 kg cow<sup>-1</sup> day<sup>-1</sup>) and animals grazing the high HM swards (23.6 kg cow<sup>-1</sup> d<sup>-1</sup>). Milk yield was greater ( $P < 0.05$ ) for animals offered 20 kg DHA (24.1 kg cow<sup>-1</sup> d<sup>-1</sup>) than those offered 16 kg DHA (23.1 kg cow<sup>-1</sup> d<sup>-1</sup>). There were no differences in milk fat or lactose concentrations between treatments. Daily herbage allowance had a significant effect ( $P < 0.01$ ) on milk protein concentration. Milk protein concentration of treatments offered 16 kg DHA was 32.5g kg<sup>-1</sup> and this increased to 33.3 g kg<sup>-1</sup> when 20 kg DHA was offered. Milk fat yield increased ( $P < 0.01$ ) as DHA increased. Milk fat yield of animals offered 16 kg DM DHA was 851.4 g cow<sup>-1</sup> d<sup>-1</sup>; this increased to 906.9 g cow<sup>-1</sup> d<sup>-1</sup> for animals offered 20 kg DM DHA. Similarly, milk protein yield increased significantly ( $P < 0.01$ ) as DHA increased from 16 kg DM (749.8 g cow<sup>-1</sup> d<sup>-1</sup>) to 20 kg DM DHA (801.7 g cow<sup>-1</sup> d<sup>-1</sup>). Milk lactose yield increased ( $P < 0.05$ ) as DHA increased. Milk lactose yield of animals offered 16 kg DM DHA was 1037.2 g cow<sup>-1</sup> d<sup>-1</sup>; this increased to 1087.0 g cow<sup>-1</sup> d<sup>-1</sup> for animals offered 20 kg DM DHA.

Table 1. Effect of herbage mass and daily herbage allowance on milk production of mid-lactation dairy cows.

HM (kg DM ha <sup>-1</sup> )	< 1600		< 2100		SED	HM	DHA	HM*DHA
DHA (kg DM cow <sup>-1</sup> day <sup>-1</sup> )	16	20	16	20				
Milk yield (kg day <sup>-1</sup> )	23.0	24.1	23.2	24.0	0.58	ns	*	ns
Milk fat content (g kg <sup>-1</sup> )	37.1	37.9	37.1	37.7	0.85	ns	ns	ns
Milk protein content (g kg <sup>-1</sup> )	32.6 <sup>ac</sup>	33.4 <sup>b</sup>	33.4 <sup>c</sup>	33.2 <sup>ab</sup>	0.38	ns	**	ns
Milk lactose content (g kg <sup>-1</sup> )	45.2	45.2	44.8	45.2	0.30	ns	ns	ns
SCM yield (kg day <sup>-1</sup> )	20.5 <sup>a</sup>	21.9 <sup>b</sup>	20.4 <sup>a</sup>	21.7 <sup>b</sup>	0.61	ns	**	ns
Bodyweight (kg)	514	521	542	526	21.4	ns	ns	ns
BCS	2.6	2.7	2.7	2.7	0.05	ns	ns	ns

<sup>a-c</sup>Means within a row with different superscripts differ ( $P < 0.05$ ); \*,  $P < 0.05$ ; \*\*,  $P < 0.01$ . ns = not significant. HM = Herbage mass; DHA = Daily herbage allowance.

There was no effect of pre-grazing herbage mass on milk composition. Increasing DHA, resulted in an increase in solids corrected milk yield (SCM). Solids corrected milk yield of animals offered 16 kg DM DHA was 20.4 kg cow<sup>-1</sup> d<sup>-1</sup>; this increased by 1.4 kg cow<sup>-1</sup> d<sup>-1</sup>

when 20 kg DHA was offered. There was no effect of treatment on bodyweight or BCS during the experimental period.

## Discussion

Mayne and Laidlaw (1999) stated that increasing herbage allowance will result in an increase in the proportion of herbage uneaten, and consequently the quality of the herbage composition and sward structure deteriorates as the season progresses. Herbage utilization increases as DHA decreases. In the current study the milk response to extra herbage was 0.25 kg kg<sup>-1</sup> offered. This is similar to the response reported by Delaby *et al.* (1999). Combellas and Hodgson (1979) reported a substantial effect of DHA on milk yield within one week of treatments being imposed. There was no effect of HM on milk production in the current study, which agrees with the findings of Wales *et al.* (1999) who reported no effect of herbage mass on animal production in mid-lactation.

## Conclusions

Management of high-yielding dairy cows at pasture presents a major challenge to dairy farmers. Rising production costs and falling farm gate product prices have induced a change in grassland management in recent years. Increasing the proportion of grazed grass in the diet of the dairy cows is a major objective on grazing dairy farms. Results from this study indicate that offering mid-lactation animals a high DHA (20 kg DM cow<sup>-1</sup> d<sup>-1</sup>) will increase milk production and milk protein concentration. Furthermore, maintaining low pre-grazing herbage masses (range: 1,200-1,600 kg DM ha<sup>-1</sup>) has been shown to maintain sward quality and maximize intake on leafy swards as the season progresses.

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# Effect and carryover effect of spring grazing access time on dairy cow performance

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## Abstract

Early turnout in spring with a limited access time to pasture will allow the grazing season to be extended. The objective of this experiment was to compare three grazing access times (4, 8 and 21 hours day<sup>-1</sup>) applied over 5 weeks (phase 1) on milk yield and composition and to evaluate the carryover effect when cows grazed fulltime, later in the grazing season (4 weeks, phase 2). Sixteen Holstein mid-lactation cows were assigned to each treatment and were supplemented with 5 kg DM of Total Mixed Ration (maize silage and soybean meal). The herbage allowance was fixed at 12 kg DM cow<sup>-1</sup> d<sup>-1</sup>) during phase 1 and 14 kg DM cow<sup>-1</sup> day<sup>-1</sup> during phase 2. During phase 1, milk yield was significantly affected by access time, 24.4, 27.0 and 28.6 kg cow<sup>-1</sup> for 4, 8 and 21 hours, respectively. These differences were fully expressed at the end of the first week and remained constant thereafter. During phase 2, the cows that previously grazed for 4 h and 8 h per day produced 3.5 and 2.0 kg day<sup>-1</sup> less, respectively, than the control group. Reducing access time over 5 weeks affects the lactation potential for cows in the second part of lactation and this carryover effect appeared to be increased with cow potential at turnout.

Keywords: dairy cow, grazing, access time, milk yield, carryover effect

## Introduction

Early turnout in spring and/or late grazing in autumn are methods of extending the grazing season and increasing the proportion of grazed grass in the annual feed budget of dairy cows. During these periods, the grass growth and the amount of grass available are often insufficient to satisfy total animal demand, and inclement weather conditions increase the risk of damage by trampling. Consequently, cows are supplemented indoors and are only allowed to access the pasture for a short period of time each day. This restricted access time has consequences on grazing behaviour, grass dry matter intake (Mattiauda *et al*, 2003; Pérez-Ramírez *et al*, 2008) and finally on milk performance. The importance of these effects seems to depend on the severity of the time restriction and on the length of time during which the restriction is imposed. The aim of the work reported in this paper was to investigate the direct effects and the carryover influence of three pasture-access times applied during five weeks in spring on dairy cow performance and pasture utilization.

## Materials and methods

This experiment was conducted at the Méjusseume farm, INRA, Le Rheu in Brittany (48.11°N, 1.71°W) in spring 2006. The three access times compared were 4 (08:30-12:30), 8 (08:30-16:30) and 21 (08:30-16:30 and 18:30-07:30) hours day<sup>-1</sup> and were applied during 5 consecutive weeks (phase 1). During the next four weeks (phase 2), all the cows had access to pasture for 21 hours day<sup>-1</sup>. The 48 experimental Holstein cows were blocked in 3 balanced groups according to parity (4 primiparous /treatment), calving date (26/11/2005 – 149 ± 38 days in milk), two weeks pre-experimental milk yield (37.0 ± 3.9 kg), milk composition



( $31.6 \pm 6.2$  and  $29.3 \pm 1.6$  g/kg fat and protein content, respectively) and bodyweight ( $643 \pm 67$  kg).

The total grazing area (12.9 ha) was split into three identical areas. The 3 groups of cows grazed separately, a new area was fenced daily, according to actual herbage mass, to offer 12.0 and 14.0 kg of DM of grass  $\text{cow}^{-1} \text{day}^{-1}$  respectively during phase 1 and 2. During the two phases, each cow received 5 kg DM of a total mixed ration (TMR) which consisted on a proportional DM weight basis, of maize silage (0.85), soybean meal (0.12) and minerals (0.03) fed after the afternoon milking. No other concentrate was fed. The herbage mass at 5 cm and sward density were measured weekly by cutting 4 strips per treatment with a motor scythe on the next day's area. The harvested grass was sampled and dried to determine the DM content and was ground prior to chemical analysis (minerals, crude protein, crude fibre content and pepsin-cellulase digestibility). The pre- and post-grazing height were determined daily on each treatment to evaluate the herbage utilization according to the methodology outlined by Delaby and Peyraud (1998). Individual milk yield was recorded daily at the two milkings. Milk fat and protein content were individually measured six times per week. The cows were weighed weekly. The offered and refused supplements were individually weighed to calculate the TMR intake by difference. The statistical analyses were performed on the individual animal data using a general linear model including the effects of lactation number, access time and pre-experimental variables as covariates. The pasture data were analysed with a model including the effect of week of measurement and access time.

Table 1. Effect and carryover effect of restricted access time at grazing on dairy cow performance and pasture utilization.

Access time (hours $\text{day}^{-1}$ )	4	8	21	RSE	<i>P</i> <
<b>Phase 1 (5 weeks)</b>					
Milk yield (kg)	24.4	27.0	28.6	1.69	0.0001
Fat yield (g)	959	1041	1041	72.7	0.0031
Protein yield (g)	668	769	810	49.3	0.0001
Fat content (g $\text{kg}^{-1}$ )	39.4	38.7	36.6	2.15	0.0025
Protein content (g $\text{kg}^{-1}$ )	27.4	28.6	28.4	0.87	0.0017
Bodyweight (kg) <sup>(1)</sup>	587	610	609	17.0	0.0007
Herbage mass (kg DM/ha)	2330	2390	2309	115.6	0.5440
Pre-grazing height (cm)	14.3	15.0	14.5	0.47	0.1390
Post-grazing height (cm)	8.5	6.9	6.1	0.53	0.0005
Grass removed (kg DM $\text{cow}^{-1} \text{d}^{-1}$ )	7.6	9.7	10.7	0.34	0.0001
<b>Phase 2 (4 weeks)</b>					
Milk yield (kg)	23.1	24.5	26.6	1.94	0.0001
Fat yield (g)	855	927	949	75.7	0.0035
Protein yield (g)	686	736	782	41.9	0.0001
Fat content (g $\text{kg}^{-1}$ )	37.1	37.9	35.8	2.39	0.0492
Protein content (g $\text{kg}^{-1}$ )	29.7	30.1	29.7	1.54	0.7188
Bodyweight (kg) <sup>(1)</sup>	626	629	622	20.0	0.6283
Herbage mass (kg DM $\text{ha}^{-1}$ )	2405	2346	1980	150.8	0.0150
Pre-grazing height (cm)	14.2	14.1	13.2	0.57	0.0934
Post-grazing height (cm)	6.7	6.8	6.8	0.55	0.9928
Grass removed (kg DM $\text{cow}^{-1} \text{d}^{-1}$ )	11.2	11.3	11.2	0.79	0.9804

RSE: Residual standard error – (1) at the end of each phase.

## Results and discussion

During phase 1, the herbage mass (2,345 kg DM  $\text{ha}^{-1}$ ) and the pre-grazing height (14.6 cm) were identical between the grazing treatments, and consequently the stocking rate was 6.45 cows  $\text{ha}^{-1}$ . The quality of grass was not affected by the treatment, and average values were 219 g  $\text{kg}^{-1}$  DM crude protein and 0.74 OM digestibility. No refusals of TMR were recorded during this phase. The post-grazing height and the grass removed were significantly

( $P < 0.0005$ ) affected by the treatments. The shorter the access time the higher the post-grazing height; there was a large effect at 4 hours access time (8.5 cm). The grass removed declined by 1 kg of DM (i.e. 77 g DM h<sup>-1</sup>) between 21 and 8 hours and declined by 2.1 kg of DM (i.e. 525 g DM h<sup>-1</sup>) between 8 and 4 hours. The dairy cows' performance was affected by the restricted access time. Compared with the fulltime grazing treatment, the restricted access at 8 hours reduced the milk yield by 1.6 kg cow<sup>-1</sup> (i.e., 0.12 kg milk h<sup>-1</sup>). The effect was greater at the 4 hours access time with a reduction of 4.2 kg of milk cow<sup>-1</sup> (0.25 kg milk h<sup>-1</sup>). The milk fat content was low at 21 hours grazing access time and increased linearly at 8 and 4 hours of access time. The milk protein content was similar at 21 and 8 hours and was lower at 4 hours of access time (-1.0 g kg<sup>-1</sup>). All these effects are in line with those described by Faverdin *et al.* (2007) in the case of restricted feeding indoors with conserved forages. The most important effect observed was when cows were grazing for only 4 hours and this was the consequence of the low feeding level due to the inability of the cows to adapt their intake behaviour to a short access time. When access time was restricted, the dairy cows tried to compensate by increasing the proportion of time spent grazing and the intake rate, but were unable to maintain the same level of grass intake (Pérez-Ramírez *et al.*, 2008). These effects were fully expressed at the end of the first week and remained constant during the last four weeks of phase 1. During phase 2, the herbage mass and the pre-grazing height were higher on the previous 4-hours-treatment paddocks, due to the higher post-grazing height observed in phase 1. For the same herbage allowance, the post-grazing height and the amount of grass removed were similar to the previous treatments (11.2 kg DM cow<sup>-1</sup> day<sup>-1</sup>). A significant carryover effect was observed on milk, fat and protein yields during the post-experimental phase. Compared with fulltime grazing, a restricted access time during 5 weeks induced, on average during the first four weeks after the end of phase 1, a lower milk yield of 2.0 and 3.5 kg, respectively for the previous 8- and 4-hours treatments. No carryover effect was observed on milk protein content. The milk fat content remained significantly lower for cows allocated to the fulltime grazing treatment during phase 1. During phase 1 and phase 2, the effect and the carryover effect of restricted access time seem greater for high yielding cows. Similar carryover effects have been described by Roche (2007) after a very low herbage allowance treatment was applied during 5 weeks in early lactation. In the case of severe feeding restriction, it is possible that the numbers of mammary secretory cells and their activity may have been altered.

## Conclusion

If a restricted access time is to be recommended, in order to avoid pasture damage, it seems preferable to limit the effect on cow performance by adjusting the amount of supplementary feed and to graze for at least 8 hours per day and, if possible, to split this access time into 2 sub-periods just after milkings to capitalize on post-milking intake motivation as this has been favourably tested by Kennedy *et al.* (unpublished) in Ireland.

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## A new Nordic structure evaluation system for diets fed to dairy cows

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### Abstract

The objective of the NorFor structure system was to establish a model for prediction of eating- (EI), ruminating- (RI) and total chewing index (CI) for feeds and total rations fed to dairy cows. The model was predicted from a Meta analysis of more than 100 published experiments including results on eating, ruminating and total chewing time by cattle fed mainly high forage diets. The EI is proportional with the neutral detergent fibre (NDF) content and a particle size factor (Size\_E) of the feed. The RI is proportional with the NDF content, a particle size factor (Size\_R) and a hardness factor of the feed. The CI is estimated as the sum of the EI and RI. The Size\_E and Size\_R values are estimated from exponential functions of the most frequent particle length. The hardness factor is estimated as a linear function of the proportion of indigestible NDF in NDF. A minimum dietary CI value of 32 minutes per kg of total DMI is considered to ensure a sufficient intake of physically effective fibre in order to ensure good ruminal fibre digestion and to prevent a low rumen pH, digestive disorders and a low milk fat content in high-yielding dairy cows.

Keywords: NorFor, NDF, particle size, chewing index, cattle

### Introduction

A minimum amount of forage fibre is essential for optimal rumen function, fibre digestion and to avoid milk fat depression and digestive disorders (Mertens, 1997). The optimal forage to concentrate ratio in diets for high-yielding dairy cows increases at decreasing content of structural fibres and particle size (PS) in forages (de Brander *et al.*, 2002). The structural dietary fibre can be chemically characterized as neutral detergent fibre (NDF). The PS has been measured by sorting particles into different sieving fractions or by measuring the particle length. The overall PS distribution has been characterized by the most frequent length value (Mo\_PL) and by the upper 95 percentile value measured by sieving (PS\_95) or from the distribution of particle-length values (PL\_95). The Mo\_PL value in chopped forage equals the theoretical chopping length (TCL). The Mo\_PL value of swallowed unchopped forage boli collected at cardia in cows during eating is about 20 mm, whereas the Mo\_PL value in faeces is about 0.7 mm (Nørgaard, 2006). The PS\_95 value of 1.18 mm in faeces from cattle fed forage diets at maintenance level is defined as the *Critical PS*. Fibrous particles larger than the *Critical PS* are termed physically effective fibre (*peNDF*), and daily time spent eating and ruminating is dependent on the intake of *peNDF* (Mertens, 1997). Chopping of forages with a theoretical chopping length (TCL) longer than 20 mm has decreased mean eating time with 30 to 50% without affecting the *peNDF* value, mean ruminating time or Mo\_PL value in swallowed boli during eating (Nørgaard, 2006). Fine chopping, grinding and pelleting of forages decrease both mean eating and mean ruminating times (de Brander *et al.*, 2002;

Mertens, 1997). The objective of this paper is to present a model for prediction of eating- (EI), ruminating- (RI) and total chewing index (CI) values for feeds and total rations fed to dairy cows in the Nordic Structure System within NorFor (NorFor\_SS).

## Materials and methods

The intention behind the NorFor\_SS is to characterize the individual feeds with an additive structure value, which ranks the feeds according to their stimulus to the mastication of particles during eating and rumination. The time spent masticating and the insensitivity of mastication is considered to perform an additive biological quantitative ranking of the physical structure values, which are closely related with the size of salivation, frequency of rumen motility and the formation of a stable flowing layer system in the reticulo-rumen. The NorFor\_SS is based on the Danish chewing index (CI) system (Nørgaard, 1996), where concentrates are given a CI value of 4 or 10 min. kg<sup>-1</sup> DM, and unchopped forages are given a CI value of 300 min kg<sup>-1</sup> crude fibre. In NorFor\_SS each feedstuff is given both an eating- (EI, min. kg<sup>-1</sup> DM), a ruminating - (RI, min. kg<sup>-1</sup> DM) and a chewing index value (CI, min. kg<sup>-1</sup>DM), which is the sum of the EI and RI values (Table 1). The models for estimation of the EI, RI and CI values are presented in Table 1. The EI value is intended to reflect the associated chewing activity during eating of a feed. EI is considered to be dependent on the PS and NDF content of the feed. The PS is transformed into a Size\_E factor value, which is estimated as an exponential function of the TCL or Mo\_PL value in the feeds. The Size\_E values range from zero in coarsely processed concentrates to 1 for unchopped forages. Ground feeds or feeds with a Mo\_PL shorter than 6 mm are given an EI value of 4 minutes kg<sup>-1</sup> DM. The EI values of forages are considered to be proportional with a proportionality factor (kEI), the Size\_E value and the NDF content. The RI values are considered to be dependent on PS, NDF content and Hardness of the structural fibres in feeds. The effect of PS on rumination is transformed into a Size\_R factor value, which is estimated as an exponential function of the TCL or Mo\_PL value of a feed and to the Mo\_PL value in faeces. The Size\_R values range from 0 for finely ground feeds to 1 for unchopped forages. Size\_R has a value of 0.9 in forage chopped at 10 mm TCL. The hardness of structural fibres is characterised by a Hardness factor (H), which ranges from 0.8 in young immature forage fibre to 1.25 in highly lignified forage legume fibres. The H value is estimated as a linear function of INDF/NDF, which is expected to reflect the lignifications of the structural fibres and associated physical power required for breaking down large particles. The RI value is proportional with a proportionality factor (kRI), Size\_R, H and the NDF content of the feed. The kEI and kRI were estimated to 50 and 100 min kg<sup>-1</sup> NDF, respectively, from a metaanalysis of 50 published treatments with cows fed concentrates and forages in a long physical form. The RI and CI values were estimated on a total mixed ration fed to seven lactating Holstein Frisian cows with a mean daily DM intake of 21.2 kg (Hymøller *et al.*, 2005; Table 2).

## Results and discussion

The RI and CI values of the total mixed ration (Table 2) were estimated to 25 and 36 minutes kg<sup>-1</sup> DM. Hymøller *et al.* (2005) recorded a mean daily ruminating and chewing time for that ration of 23 ± 2 and 42 ± 3.3 minutes kg<sup>-1</sup> DM, respectively. The estimated CI value of 36 is higher than the recommended NorFor minimum of 32 min. kg<sup>-1</sup> DM (Gustafsson *et al.*, 2007) and than the results by Sudweeks *et al.* (1981). The higher observed chewing time of 42 minutes kg<sup>-1</sup> DM might be explained by difficulties in discrimination of jaw movements during eating and licking.



Table 1. Model for prediction of chewing index values in feeds for cattle.

Term	Abreaction	Unit	Estimation
Mode particle length	Mo_PL	mm	Image analysis
Chopping length <sup>a</sup>	TCL <sup>a</sup>	mm	Position of chopper
Structural fibre	NDF	kg kg <sup>-1</sup> DM	Chemical analysis
Indigestible NDF	INDF	kg kg <sup>-1</sup> DM	In sacco incubation
PS <sup>b</sup> factor eating	Size_E	0 < Size_E ≤ 1	$1 - 0.52 \times e^{-0.078 \times \text{TCL}}$
PS <sup>b</sup> factor ruminating	Size_R	0 < Size_R ≤ 1	$1 - e^{-0.173 \times (\text{TCL}/0.7-1)}$
Hardness factor	H	0.8 < H ≤ 1.25	0.75 + INDF/NDF
Eating index	EI <sup>c</sup>	Min. kg <sup>-1</sup> DM	50 x NDF x Size_E
Ruminating index	RI	Min. kg <sup>-1</sup> DM	100 x NDF x Size_R x H
Chewing index	CI	Min. kg <sup>-1</sup> DM	EI + RI

a) Theoretical chopping length equals the Mo\_PL value. b) PS: Particle size. c) EI = 4 min. kg<sup>-1</sup> DM, e.g. finely or coarsely ground feeds.

Table 2. Estimated chewing index values on a mixed ration of ground, pelleted dried beet pulp (DBP), rape seed meal (RSM), rolled barley, grass silage (GS) and early cut whole-crop maize silage (MS) with a high INDF value (Hymøller *et al.*, 2005).

Property	Unit	DBP	RSM	Barley	GS	MS
NDF	g kg <sup>-1</sup> DM	377	211	166	363	462
TCL <sup>a</sup>	mm	2 <sup>a</sup>	3 <sup>a</sup>	6	6.2 <sup>c</sup>	9.3 <sup>c</sup>
INDF <sup>a</sup>	g kg <sup>-1</sup> DM	31	94	31	51	106
Size_E <sup>a</sup>	Numeric				0.68	0.75
Size_R <sup>a</sup>	Numeric	0.27	0.43	0.73	0.74	0.88
H <sup>a</sup>	Numeric	0.83	1.2	0.93	0.89	0.98
EI <sup>a</sup>	min. kg <sup>-1</sup> DM	4	4	4	12	17
RI <sup>a</sup>	min. kg <sup>-1</sup> DM	8.6	11	11	24	40
CI <sup>a</sup>	min. kg <sup>-1</sup> DM	13	15	15	36	57
Proportion <sup>b</sup>	g kg <sup>-1</sup> DM	79	200	121	195	395

a) see Table 1, b) including 10 g kg<sup>-1</sup> DM mineral mixture c) Measured Mo\_PL value

## Conclusions

A new Nordic system for evaluation of the physical structure in diets for dairy cows by means of a model for predicting an eating- (EI), a ruminating (RI) and a total chewing index value (CI) is described.

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# Estimation of the digestion rate and digestibility of forages using *in-vitro* gas production technique

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## Abstract

Digestibility is the major factor constraining the intake and production potential of forages. Forage digestibility depends on the fibre (NDF) concentration and quality, i.e. the rate of digestion ( $k_d$ ) of potentially digestible fibre (pdNDF). We used 90 samples from grass, legume and whole-crop cereal silages. The feeds were grown in Finland and harvested at different stages of maturity. *In vivo* organic matter digestibility (OMD) of the silages was determined by total faecal collection method with sheep fed at maintenance level of intake. The potential extent of NDF digestion was determined by a 12-d ruminal *in situ* incubation. Isolated silage NDF was incubated for 4 d in the presence of rumen fluid and buffer to produce the cumulative gas production curves. The  $k_d$  of pdNDF measured by *in vitro* gas production was on average 0.061 per h, and it decreased with progressing maturity of forages. The OMD could be predicted from *in vitro* gas production accurately and precisely (RMSE = 0.0099,  $R^2 = 0.968$ ). The *in vitro* gas production technique shows potential in characterizing forage quality and providing information of forage  $k_d$  for dynamic mechanistic feed evaluation models.

Keywords: silage, modelling, rate of digestion, fibre

## Introduction

Digestibility is the most important parameter affecting the production potential of forages. Digestibility determines the available energy concentration of the feeds but also affects the intake of forages (Huhtanen *et al.*, 2007). The variation of digestibility in forages is great due to differences in e.g. species and stage of maturity, which emphasizes the importance of digestibility determination for practical ration formulation.

Forage digestibility depends on the fibre (neutral detergent fibre, NDF) concentration and quality. Available methods for predicting digestibility of feeds include *in vitro* digestibility with rumen bacteria or enzymes, *in situ* incubation in nylon bags and near infrared reflectance spectroscopy. All these methods have close relationship with digestibility (Huhtanen *et al.*, 2006). The objective of this work was to find out if the *in vitro* gas production method can be used to estimate the rate of digestion ( $k_d$ ) of potentially digestible fibre (pdNDF). Accurate and precise estimates of  $k_d$  are needed in dynamic mechanistic modelling of feed digestion in the rumen. We used the same data set as Huhtanen *et al.* (2006) including silages made from primary growth and regrowth grass, legumes and whole crop cereals.

## Materials and methods

The data set comprised primary growth grass (PG,  $n = 33$ ), regrowth grass (RG,  $n = 27$ ), legume (L,  $n = 23$ ) and whole-crop cereal (WCC,  $n = 7$ ) silages with OMD values determined

*in vivo*. The feeds were grown in Finland over a 10-year period and harvested at different stages of maturity. Methods in silage production, digestibility trials and chemical and *in vitro* analyses have been described by Huhtanen *et al.* (2006). The OMD of the silages was determined by total faecal collection method in sheep fed at maintenance level of intake. The indigestible NDF concentration (iNDF) was determined by a 12-d ruminal *in situ* incubation using polyester bags with a small (6 or 17  $\mu$ m) pore size.

*In vitro* gas production measurements were made by an automated system. Isolated silage NDF samples in three replicates were incubated for 3–4 d in the presence of rumen fluid and buffer to produce cumulative gas production curves. A two-pool Gompertz model was fitted to the cumulative gas curves and the Gompertz model parameters were then used in a two-compartment mechanistic rumen model to predict the digestibility of pdNDF as described by Huhtanen *et al.* (2008). The  $k_d$  of pdNDF ( $k_{d-GAS}$ ) was calculated backwards from digestibility of pdNDF as described by Huhtanen *et al.* (2006). The digested neutral detergent solubles (NDS) were calculated using the Lucas equation approach (see Huhtanen *et al.*, 2006) and digested OM as digested NDF + digested NDS.

The relationships between the *in vivo* values and the laboratory measurements were examined using regression analysis and the SAS Mixed model approach. The *in vivo* digestibility trials ( $n = 23$ ) were used as the random variable, and using them will correct for the annual changes in feed characteristics, experimental procedures and variability between the *in vitro* gas production incubations.

## Results and discussion

The average OMD (g/g), and NDF and iNDF concentrations (g/kg DM) of PG, RG, L and WCC datasets were 0.733, 0.694, 0.707, and 0.686; 568, 533, 369 and 432 and 79, 106, 109 and 119, respectively. The data sets were almost identical to those used by Huhtanen *et al.* (2006). The  $k_d$  of pdNDF measured by *in vitro* gas production method was on average 0.059 (SD = 0.0109), 0.056 (SD = 0.0089), 0.068 (SD = 0.0076), and 0.064 (SD = 0.022) per h for PG, RG, L and WCC silages, respectively. Progressing maturity decreased the  $k_d$  of PG, RG and L ( $R^2$  between iNDF concentration and  $k_d = 0.811$ , 0.953 and 0.892), but for WCC, the effect was not very clear ( $R^2 = 0.220$ ).

Table 1 shows the predictions of *in vivo* OMD for the different data sets and for all data combined. The iNDF concentration was a very good predictor of OMD as previously presented by Huhtanen *et al.* (2006). The relationship between OMD and  $k_d$  of pdNDF was not so close, but when  $k_d$  was used as a second variable with iNDF in the regression equation, the residual mean squared error was improved especially for PG silages (0.0087 vs. 0.0063 g/g). This demonstrates that the  $k_d$  values produced by the *in vitro* gas production technique were biologically meaningful. The relationship between OMD produced *in vivo* vs. *in vitro* gas production were very close (Table 1.) There seems to be differences between the forage types because using the forage type specific relationships produced better predictions than all data combined. The WCC silages were an exception, and the data should be extended to increase the validity of the results concerning WCC.



Table 1. Prediction of *in vivo* organic matter digestibility (g/g) from indigestible NDF (iNDF, g/kg DM) concentration, rate of degradation of potentially digestible NDF ( $k_{d-GAS}$ , 1/h) and OMD derived from the *in vitro* gas production measurements (OMD<sub>GAS</sub>) using mixed model regression analysis for primary growth grass (PG, n = 33), regrowth grass (RG, n = 27), legume (L, n = 23) and whole crop cereal (WCC, n = 7) silages and all data combined.

Species	X <sub>1</sub>	X <sub>2</sub>	Intercept	X <sub>1</sub>	X <sub>2</sub>	RMSE <sup>1</sup>	Adj. R <sup>2</sup>
PG	iNDF		0.851	-0.00149		0.0087	0.978
RG	iNDF		0.843	-0.00135		0.0073	0.971
L	iNDF		0.834	-0.00110		0.0128	0.954
WCC	iNDF		0.867	-0.00152		0.0082	0.970
ALL	iNDF		0.835	0.00125		0.218	0.840
PG	$k_{d-GAS}$		0.401	5.45		0.0212	0.886
RG	$k_{d-GAS}$		0.365	6.03		0.0118	0.954
L	$k_{d-GAS}$		0.233	6.77		0.0253	0.803
WCC	$k_{d-GAS}$		0.651	0.35		0.0108	0.273
ALL	$k_{d-GAS}$		0.526	3.01		0.0462	0.280
PG	iNDF	$k_{d-GAS}$	0.749	-0.00119	1.29	0.0063	0.989
RG	iNDF	$k_{d-GAS}$	0.725	-0.00108	1.59	0.0072	0.972
L	iNDF	$k_{d-GAS}$	0.726	-0.00093	1.30	0.0145	0.937
WCC	iNDF	$k_{d-GAS}$	0.866	-0.00152	0.01	0.0092	0.962
ALL	iNDF	$k_{d-GAS}$	0.762	-0.00110	0.95	0.0123	0.948
PG	OMD <sub>gas</sub>		0.0351	0.963		0.0058	0.990
RG	OMD <sub>gas</sub>		0.0281	0.998		0.0077	0.943
L	OMD <sub>gas</sub>		0.0375	0.951		0.0124	0.956
WCC	OMD <sub>gas</sub>		0.3707	0.436		0.0174	0.737
ALL	OMD <sub>gas</sub>		0.0552	0.932		0.0099	0.968

<sup>1</sup>RMSE = Residual mean squared error.

## Conclusions

The *in vitro* gas production technique shows potential in characterizing forage quality and providing information of forage  $k_d$  for mechanistic dynamic feed evaluation models.

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# Effect of grass silage chop length on intake and milk production by dairy cows

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## Abstract

Roundbale grass silage harvested at an early (D-value 757 g kg<sup>-1</sup> DM) or normal (D-value 696 g kg<sup>-1</sup> DM) stage of maturity was used to study the effect of harvesting time, chop length and their interaction on silage intake by dairy cows. Six early lactating Norwegian Red (NRF) dairy cows were used in a 6 x 6 Latin square design with 3-week periods. The two silages were fed unchopped (170 mm median particle length (MPL)), coarsely chopped to 55 mm MPL, or finely chopped to 24 mm MPL. Cows were fed silage *ad libitum* and supplemented with 6 kg concentrate. Silage intake increased only slightly after coarse chopping (+ 0.4 kg DM), but significantly after fine chopping (+ 1.3 kg DM), compared with unchopped silage. No interaction between harvesting time and chop length was found. Milk yield response was low (+ 0.5 kg ECM for finely chopped silage) compared with the increased intake. Body weight changes were not recorded due to the short periods. No indications of lack of physical structure were found when well preserved, highly digestible, low-DM grass silage was chopped to 24 mm prior to feeding.

Keywords: chop length, cows, early harvested silage, intake

## Introduction

Previous studies have indicated that grass silage intake and milk yield is higher when silage is chopped to 10-20 mm compared with 70-150 mm (Dulphy, 1980). Most studies on different chop lengths, however, are performed using different harvesting equipment. This makes it difficult to separate the indirect effect on intake caused by different silage fermentation qualities and the direct effect of different chop lengths. The effect of chop length increases with increased silage proportion in the diet (Murdoch, 1965). Very finely chopped silage (5 mm theoretical chop length) fed in a high concentrate (0.65) diet reduced intake and milk yield compared with longer chop length (10 mm) or less concentrates (0.35), probably due to lack of physical structure (Beauchemin *et al.*, 1994). The objective of the present study was to evaluate the effect of chop length of grass silage harvested at two stages of maturity.

## Materials and methods

The primary growth of a sward consisting of (kg<sup>-1</sup> DM) 636 ± 141 g timothy (*Phleum pratense*), 108 ± 44 g meadow fescue (*Festuca pratensis*) and 255 ± 153 g red clover (*Trifolium pratense*) was harvested at Ås, Norway (60°N, 11°E) in 2005 at two stages of maturity: (1) Early (E), 1-2 June, mean stage by weight (MSW) of timothy = 2.44 (Moore *et al.*, 1991), D-value = 757 g kg<sup>-1</sup> DM, 200 g CP kg<sup>-1</sup> DM, 424 g NDF kg<sup>-1</sup> DM; and (2) Normal (N), 16 June, MSW = 3.01, D-value = 696 g kg<sup>-1</sup> DM, 150 g CP kg<sup>-1</sup> DM, 516 g NDF kg<sup>-1</sup> DM. The herbage was baled using an Orkel GP 1260 roundbaler with 20 fixed knives, applied 4.3 (E) and 3.1 (N) L Mg<sup>-1</sup> GrasAAT Lacto (780 g formic acid kg<sup>-1</sup>) and wrapped in 6 layers of 0.025 mm thick and 750 mm wide white plastic film (Rani Plast OY). Both E and N silage was fed long (L) with no further chopping; coarsely chopped (C) using a Serigstad 1200 RBK roundbale chopper; or finely chopped (F) (treated as C plus further chopping using epple Blasius 940). The six silages were fed *ad libitum* to six Norwegian Red

dairy cows ( $27 \pm 9$  d.i.m.) in 2<sup>nd</sup> to 6<sup>th</sup> lactation in a 6 x 6 Latin square design with six three-week periods. The cows were supplemented with 6 kg d<sup>-1</sup> of concentrate (kg<sup>-1</sup>: 120 g rape seed cake, 100 g soypass, 380 g barley, 248 g oats, 60 g wheat bran, 70 g molasses and 22 g lignobond, minerals and vitamins), and 150 g d<sup>-1</sup> of a mineral and vitamin supplement (kg<sup>-1</sup>: 110 g Ca, 70 g P, 65 g Mg, 90 g Na).

## Results

Median chop lengths measured by sorting by hand were for silage E: 184 mm (L), 57 mm (C) and 24 mm (F), and for silage N: 153 mm (L), 53 mm (C) and 23 mm (F). Chemical composition of silages and concentrate is given in Table 1. Silage E and N contained (kg<sup>-1</sup> DM) 54 and 73 g lactic acid, 12 and 11 g formic acid, 16 and 17 g acetic acid, 2.1 and 1.5 g propionic acid, 0.2 and 0.0 g butyric acid and 14 and 23 g ethanol, respectively. Ammonium N concentration corrected for additive-derived N was 55 and 101 g kg<sup>-1</sup> N, and pH was 4.23 and 4.16 for silage E and N, respectively. Both silages were well preserved, although ethanol and ammonium concentrations preferably could have been lower.

Table 1. Composition of silage and concentrate.

	Early harvested silage	Normal harvested silage	Concentrate
DM, g kg <sup>-1</sup>	199	221	879
<u>g kg<sup>-1</sup> DM:</u>			
OM	925	922	928
CP	196	141	178
NDF	414	510	227
Fat	44	30	39
Starch plus sugar			341
Sugar (monosacharides + sucrose)	65	20	

Table 2. Feed intake and milk production.

	Early harvested silage			Normal harvested silage			s.e.m.	Effect (P)		
	Long	Coarse	Fine	Long	Coarse	Fine		Time <sup>1</sup>	Chop <sup>2</sup>	T x C <sup>3</sup>
Silage, kg DM	16.7	17.1	17.9	14.8	15.2	16.3	0.33	<0.001	0.002	ns
Concentrate, kg DM	5.04	5.26	5.26	5.25	5.27	5.27				
Total ration, kg DM	21.7	22.3	23.2	20.0	20.5	21.5	0.35	<0.001	0.002	ns
Silage DM, g kg <sup>-1</sup> BW	27.5	28.1	29.4	24.4	25.1	26.7	0.54	<0.001	0.003	ns
Total DM, g kg <sup>-1</sup> BW	35.8	36.8	38.1	33.1	33.8	35.4	0.58	<0.001	0.002	ns
NDF, g kg <sup>-1</sup> BW	13.3	13.6	14.1	14.4	14.7	15.6	0.28	<0.001	0.004	ns
Milk, kg	31.8	30.9	32.2	29.6	30.0	30.1	0.80	0.01	ns	ns
ECM, kg	33.2	32.1	33.5	29.9	30.5	30.6	0.89	0.002	ns	ns
Fat, g kg <sup>-1</sup>	43.8	43.6	43.4	42.1	42.8	42.4	0.7	0.06	ns	ns
Protein, g kg <sup>-1</sup>	32.6	33.2	33.3	31.2	31.1	31.5	0.4	<0.001	ns	ns
Lactose, g kg <sup>-1</sup>	47.0	46.5	46.9	47.5	47.4	46.7	0.3	ns <sup>4</sup>	ns	ns
Fat, g	1392	1345	1390	1239	1275	1277	43	0.004	ns	ns
Protein, g	1033	1014	1063	919	923	943	24	<0.001	ns	ns
Lactose, g	1496	1438	1513	1404	1418	1406	45	0.06	ns	ns
Urea, mM	4.91	4.95	4.80	4.58	4.42	4.55	0.20	0.03	ns	ns
FFA, meq L <sup>-1</sup>	0.79	0.85	0.92	1.10	0.82	1.10	0.13	ns	ns	ns
N in milk (N in feed) <sup>-1</sup>	0.244	0.235	0.238	0.299	0.294	0.287	0.005	<0.001	ns	ns

<sup>1</sup>Harvesting time <sup>2</sup>Chop length <sup>3</sup>Harvesting time x chop length interaction <sup>4</sup>ns:  $P \geq 0.1$ .

Dry matter intake was higher of silage E than N, but NDF intake was highest of silage N (Table 2). Fine chopping increased intake significantly, and no silage by chop length interaction was detected. Milk yield was significantly higher when cows were fed silage E than silage N, with increased concentration of fat, protein and milk urea. The utilization of feed N for milk production was lower for silage E than N.

No significant effects of chop length on yield parameters were found, but numerically milk yield was 0.5 kg higher on F than on L, with 0.5 g kg<sup>-1</sup> higher protein concentration.

## Discussion

The chop length reduction from approximately 170 mm (L) to 24 mm (F) increased intake by 0.072 for silage E and 0.101 for silage N, in diets consisting of approximately 0.75 silage and 0.25 concentrate, in agreement with previous studies (Randby, 2005, 2006). Chop length reduction to 55 mm (C), however, did not increase intake significantly, in agreement with Toivonen and Heikkilä (2005). Total ration digestibility was high, and not influenced by chop length, and neither was rumen liquid composition influenced by chop length (Garmo *et al.*, 2008). The small milk yield response to the increased intake of silage F was therefore most probably due to increased body fat deposition, and not a result of poor energy utilization of finely chopped silage.

## Conclusions

Compared with long silage (170 mm chop length), the effect on intake of coarsely chopped silage (55 mm) was small, whereas the effect of finely chopped silage (24 mm) was great both for silage harvested at an early and at a normal stage of maturity.

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# The impact of cow genotype on the profitability of grassland-based beef production in Ireland

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## Abstract

In Ireland, replacement breeding heifers for suckler beef production are increasingly being sourced from within the suckler herd resulting in an upgrading of many suckler beef cows to pure breeds and a subsequent reduction in cow milk production, pre-weaning growth rates and carcass weights of suckler progeny. The impact of this breeding policy on the profitability of grassland-based suckler beef production was investigated using a bioeconomic simulation model, the Teagasc Farm Systems Model. Furthermore, any interaction with replacement rate was explored. Results indicate that where heifer replacements are sourced from the dairy herd profitability is greater than where replacements are retained from within the suckler herd and that increasing replacement rate had a modest impact on profitability.

Keywords: farm profitability, replacement rate, simulation model, suckler beef systems, suckler cow genotype

## Introduction

Due to the increased proportion of suckler beef cows in the Irish national cow herd and the increased use of high production Holstein strains in the dairy herd, beef cross heifers from the dairy herd suitable for suckler beef production is becoming increasingly limited. The sourcing of replacement heifers from outside of suckler herds is also undesirable from a herd health perspective. The result has been an increased retention of replacements from within the suckler herd. This breeding policy has important implications for cow energy requirements (McGee *et al.*, 2005a) and milk production potential (McGee *et al.*, 2005b). As a consequence of lower milk intake and loss of heterosis, progeny weaning weights (McGee *et al.*, 2005b) and carcass weights (Drennan *et al.*, 2005) are reduced. The objective of this study was to examine the effect of beef cow genotype and replacement policy on the profitability of grassland-based suckler beef systems. Since a change in genotype may have implications for the fertility of the suckler herd, the impact of replacement rate was also explored.

## Materials and methods

The Teagasc Farm Systems Model (TFSM) is a bioeconomic model of Irish grassland-based suckler beef systems. The model is structured as a whole farm budgetary simulation model formulated in an Excel spreadsheet. The primary farm activities of a typical Irish suckler beef farm are specified. Feed intake requirements for each animal category are calculated. The forage sub-model specifies the herbage availability throughout the year and when coupled with the animal sub-model, the stocking rate is determined.

In this study the effect of suckler cow genotype, replacement policy and replacement rate on the profitability of suckler beef systems was examined. The systems analysed were spring-calving with the objective of maximising grazed grass as a proportion of the total feed budget and minimising the feeding of concentrates. Two beef cow genotypes were examined; beef cross heifers from the dairy herd (Beef-Friesian; BF) and replacements retained from within the suckler herd (Charolais; CH). The impact of beef cow breed was modelled by matching the production coefficients of interest to breed type. The coefficients modelled were dam milk



production (McGee *et al.*, 2005b) and liveweight changes (McGee *et al.*, 2005a) and progeny pre-weaning (McGee *et al.*, 2005b) and post-weaning performance and carcass characteristics (Drennan *et al.*, 2005). Replacements are assumed to be purchased as calves in the BF scenarios and retained from within the herd in the CH scenarios. Two replacement rates, 20 percent (standard; S) and 33 percent (high; H) were also examined. A number of coefficients within the model reflected the varying effects of altering the replacement rate of the herd. For primiparous cows, birthweight and liveweight gain of progeny were reduced whilst calving difficulties and calf mortality at calving were increased compared to mature cows. Liveweight and milk yield post-partum were both reduced substantially for primiparous cows compared to mature cows. The model assumptions used are presented in Table 1. In all scenarios, stocking rate and inorganic nitrogen application rate was limited by the Nitrates Directive (Department of the Environment, Heritage and Local Government, 2006). In the suckler-to-weanling scenarios progeny were sold at 8 months of age whereas in the suckler-to-beef scenarios steers and heifers were sold at 24 and 20 months of age respectively.

Table 1. Assumptions used in model scenarios.

Area farmed	50ha
Mean calving date	24 March
Grass silage harvest date	5 June
Grass silage quality (dry matter digestibility)	676 g kg <sup>-1</sup>
Weanling steer price	€2.10 kg <sup>-1</sup> liveweight
Weanling heifer price	€1.85 kg <sup>-1</sup> liveweight
Beef price (U3 steer)	302c kg <sup>-1</sup> carcass
Beef price (R3 heifer)	299c kg <sup>-1</sup> carcass
Finishing concentrate price	€265 Mg <sup>-1</sup>
CAN price	€250 Mg <sup>-1</sup>
Replacement calf price	€150 head <sup>-1</sup>

## Results and discussion

The results for the calf-to-weanling systems are presented in Table 2. Stocking rates were similar for all scenarios. However, since replacements were sourced from within the herd in the CH scenarios, the numbers of weanlings sold were fewer. Weaning weights were also lower in the CH scenarios and hence gross output and net profit was lower than for the BF scenarios. Higher replacement rates resulted in lower cow numbers, fewer calves sold and a modest reduction in weaning weights. However, the additional output in the form of cull cows overcame any potential reduction in gross output. Therefore, the net margin for the standard and high replacement rate scenarios were similar.

Table 2. Technical and financial results of the suckler-to-weanling scenarios

Scenario <sup>1</sup>	BF-S	CH-S	BF-H	CH-H
Stocking rate (LU ha <sup>-1</sup> )	1.87	1.89	1.78	1.82
Cows calving (head)	94.4	97.2	89.7	93.9
Weanlings sold (head)	88.8	73.6	84.3	61.9
Weaning weight (kg head <sup>-1</sup> ) <sup>2</sup>	303	283	298	279
Financial results (€ ha <sup>-1</sup> )				
Gross output	1,400	1,296	1,409	1,300
Variable costs	509	481	524	484
Gross margin	891	815	885	816
Net margin	437	354	435	356

<sup>1</sup>BF = Beef-Friesian genotype; C = Charolais genotype; S = standard replacement rate (20%); H = high replacement rate (33%).

<sup>2</sup>Average of steer and heifer weanlings.

The results of the calf-to-beef scenarios are presented in Table 3. As for the weanling systems, stocking rates were similar but greater animal sales and carcass weights in the BF scenarios relative to the CH scenarios resulted in higher profit in the BF scenarios particularly in the high replacement rate scenarios where net profit was 14 percent greater than the CH scenario. Similar to the suckler-to-weanling scenario, increasing replacement rate had a modest effect on net margin.

Table 3. Technical and financial results of the suckler-to-beef scenarios.

Scenario <sup>1</sup>	BF-S	CH-S	BF-H	CH-H
Stocking rate (LU ha <sup>-1</sup> )	1.58	1.64	1.53	1.61
Cows calving (head)	59.5	64.8	57.7	65.8
Weanlings sold (head)	55.3	48.4	53.5	42.8
Weanling weight (kg head <sup>-1</sup> ) <sup>2</sup>	336	329	333	327
Financial results (€ ha <sup>-1</sup> )				
Gross output	1,396	1,345	1,397	1,340
Variable costs	539	521	543	524
Gross margin	858	824	854	816
Net margin	421	379	421	369

<sup>1</sup>BF = Beef x Friesian genotype; C = Charolais genotype; S = standard replacement rate (20%); H = high replacement rate (33%).

<sup>2</sup>Average of steers and heifers.

## Conclusions

It is likely that sourcing heifer replacements from within the suckler herd will predominate as the availability of suitable beef cross heifers from the dairy herd becomes increasingly limited. However, this study demonstrated that for suckler farmers, sourcing replacements from the dairy herd result in greater profitability due to higher output; a consequence of heavier liveweights at sale and greater numbers of animals sold. Heavier weights obtained for the BF genotype are a result of higher pre-weaning weights due to the greater milk production of this genotype. Despite the CH genotype having a higher killing out percentage, the lower finishing liveweight is not entirely mitigated and carcass weights remain lower than the BF genotype. The lower animal output in the CH scenarios is a result of the replacement policy employed; replacing from within the herd reduces potential animal sales. There was a modest effect of replacement rate on net margin. In general, the reduction in progeny sales was largely offset by the increase in cull cow sales.

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# Kura clover and reed canarygrass: a versatile mixture for dairy production in northern environments

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## Abstract

Mixtures of kura clover (*Trifolium ambiguum* Bieb.) with reed canarygrass (*Phalaris arundinacea* L.) persist indefinitely in Wisconsin, USA. Two experiments were conducted to test the performance of dairy cattle on these mixtures. In Experiment 1, Holstein cows, managed under a rotational grazing system, grazed binary mixtures of kura clover with reed canarygrass, tall fescue (*Festuca arundinacea* Schreb.) or Kentucky bluegrass (*Poa pratensis* L.) with a daily pasture allowance of 23 kg DM. All cows were offered 7.8 kg DM d<sup>-1</sup> corn-soybean meal supplement. Milk production was similar (28.1 kg d<sup>-1</sup>) for the three pastures. In Experiment 2, DM intake was slightly higher (24.2 kg vs 22.9 kg d<sup>-1</sup>) and milk production similar (34.6 kg vs 33.4 kg d<sup>-1</sup>) for cows fed alfalfa (*Medicago sativa* L.) or kura clover-reed canarygrass silage. Although kura clover-reed canarygrass diets were higher in NDF than alfalfa silage diets, alfalfa fibre was less digestible and cows produced similarly when fed diets containing approximately 60% forage. The mixture of kura clover and reed canarygrass is a promising alternative pasture and silage crop for northern environments.

Keywords: Kura clover, reed canarygrass, silage, pasture, dairy

## Introduction

The mixture of kura clover (*Trifolium ambiguum* Bieb.) and reed canarygrass (*Phalaris arundinacea* L.) has proven to be a persistent and high yielding forage source in the northern USA (Contreras *et al.*, 2008). Both kura clover and reed canarygrass have rhizome systems which contribute to persistence and grazing tolerance. The nutritive value of kura clover is greater than most other forage legumes adapted to the region, resulting in mixtures with grass having fibre and protein concentrations similar to alfalfa (Zemenchik *et al.*, 2002; Contreras *et al.*, 2008). Mixtures of reed canarygrass and kura clover have been ensiled in laboratory silos resulting in pH and volatile fatty acid profiles indicative of excellent fermentation (Contreras *et al.*, 2008). Agronomic performance and laboratory assays of feeding value suggest that the mixture of kura clover and reed canarygrass could be an excellent forage source, but data on dairy cow performance are lacking. Experiments were conducted to (1) compare milk production by dairy cows grazing three grass-kura clover mixed pastures; and (2) compare performance of lactating dairy cows fed a total mixed ration containing equal DM from either alfalfa silage or kura clover-reed canarygrass silage as the sole source of forage in the diet.

## Materials and methods

In Experiment 1, three grass-kura clover mixed pastures with the same daily allowance were compared for milk production by dairy cows. The experimental design was a 3 x 3 Latin square replicated ten times with three cows per square, three treatments (grass-kura clover mixtures), and three periods. Each period was 14 d (11 d of adaptation, and 3 d of sampling). Milk production was electronically recorded and milk samples were collected for the last 3 d of each period. Pastures consisted of binary mixtures of kura clover (cv. Endura) with either

the low alkaloid reed canarygrass (cv. Palaton) (RCK), an endophyte-free tall fescue (*Festuca arundinacea* Schreb.) cv. Select (TFK), or Kentucky bluegrass (*Poa pratensis* L.) cv. Park (KBK). Thirty multiparous Holstein cows, under a rotational grazing management system, were used to compare milk production and composition across pastures. Cows grazed the paddocks for 19 h d<sup>-1</sup> and were offered approximately 23 kg DM d<sup>-1</sup> by adjusting paddock size daily. Cows were offered 7.8 kg DM d<sup>-1</sup> of corn-soybean meal supplement.

In Experiment 2, twenty lactating Holstein cows were used in a cross-over design to compare milk production from diets containing kura clover-reed canarygrass silage (mixture silage) or alfalfa silage. The mixture was approximately 0.40 kura clover and 0.60 reed canarygrass. Fermentation product profiles indicated silages underwent normal fermentation. Treatments were total mixed rations formulated with either: (1) 0.57 of total DM as alfalfa silage, or (2) 0.57 of total DM as mixture silage. Experimental periods were 28 d, with the first 14 d for diet adaptation and the last 14 d for measurement of intake and milk production. Milk samples were collected four times during the last 7 d of each period.

## Results and discussion

In Experiment 1, kura clover comprised approximately 0.37 of the RCK, 0.45 of the TFK and 0.75 of the KBK pastures (Table 1). The three pastures had similar available herbage mass (2791, 2838 and 3236 kg DM ha<sup>-1</sup> for RCK, TFK and KBK, respectively) but differed in sward height and density. They also differed in neutral detergent fibre (NDF) contents: 419, 405, and 331 g kg<sup>-1</sup> for RCK, TFK and KBK, respectively. Milk yield was 27.0, 27.2, and 29.1 kg d<sup>-1</sup> for cows grazing RCK, TFK, and KBK pastures, respectively. Milk yield, milk protein concentration and yield, and milk urea nitrogen were all slightly greater for cattle grazing KBK pasture, because of the high legume content of this mixture. Differences in sward structure and density, as well as legume proportion, among pasture types could account for slight differences in intake and milk production observed in this study. Dry matter intake is associated with the cow's opportunity to graze selectively, a function of pasture heterogeneity and sward structure. Slightly greater production was associated with greater presence and consumption of kura clover in Kentucky bluegrass-kura clover pasture. Cattle were observed to select against stems and pseudostems of reed canarygrass. Although differences ( $P < 0.05$ ) in milk production among pastures were detected, these differences were not large. Full season pasture production was not measured in this experiment, but would be expected to be substantially lower for KBK than for the other two pastures (Zemenchik *et al.*, 2002).

Table 1. Characteristics of pasture grazed by lactating dairy cows allocated in binary mixtures of kura clover with reed canarygrass (RCK), tall fescue (TFK) or Kentucky bluegrass (KBK).

	Treatments		
	RCK	TFK	KBK
Herbage available, kg DM ha <sup>-1</sup>	2791	2838	3236
Height, cm	58	42	30
Density, kg DM m <sup>-3</sup>	0.48 <sup>b</sup>	0.67 <sup>b</sup>	1.08 <sup>a</sup>
Proportion grass:legume	63:37 <sup>b</sup>	55:45 <sup>b</sup>	25:75 <sup>a</sup>
NDF (g kg <sup>-1</sup> DM)	419 <sup>a</sup>	415 <sup>a</sup>	331 <sup>b</sup>
ADF (g kg <sup>-1</sup> DM)	237	239	212
CP (g kg <sup>-1</sup> DM)	229 <sup>a</sup>	207 <sup>b</sup>	243 <sup>a</sup>

<sup>a,b</sup>Values in the same rows with different superscripts differ ( $P < 0.05$ ).

In Experiment 2, NDF concentrations of the mixture and alfalfa silage were 473 and 374 g kg<sup>-1</sup>, respectively (Table 2). The nutritive value of the alfalfa used in this experiment was extremely high, and better than that typically fed in Wisconsin. Dry matter intake was greater

(24.2 vs. 22.8 kg d<sup>-1</sup>) but milk production was similar (34.6 vs. 33.4 kg d<sup>-1</sup>) (Table 3) for cows fed alfalfa silage vs. mixture silage. Milk fat (36 g kg<sup>-1</sup>) and milk true protein (31.7 g kg<sup>-1</sup>) did not differ due to treatment. Milk urea nitrogen content was higher when cows consumed alfalfa silage than mixture silage; this being associated with greater CP in alfalfa. Cows consumed more NDF from forage (6.5 kg vs 5.4 kg) and more digestible NDF from forage (4.2 kg vs 2.4 kg) when fed mixture diets than alfalfa diets. Although mixtures were higher in NDF than alfalfa, NDF digestibility was much greater in mixtures. Cows produced similar amounts of milk when fed diets containing approximately 60% alfalfa or kura clover-reed canarygrass silage.

Table 2. Nutrient composition and intake of alfalfa and reed canarygrass-kura silages.

	Alfalfa Silage	Kura-Reed Silage
DM, g kg <sup>-1</sup> fresh weight	363	429
CP, g kg <sup>-1</sup> DM	220	161
ADF, g kg <sup>-1</sup> DM	326	340
NDF, g kg <sup>-1</sup> DM	374	473
NDFD <sup>1</sup> , g kg <sup>-1</sup> NDF	445	649
Lignin, g kg <sup>-1</sup> DM	66	39
DM Intake, kg d <sup>-1</sup>	24.2	22.8

<sup>1</sup>NDFD = NDF digestibility, determined from in vitro fermentation for 48 h.

Table 3. Comparison of alfalfa silage (AS) and kura clover-reed canarygrass silage (KRS) diets on milk yield and milk composition.

	AS Diet		KRS Diet		P-value
	Mean	SEM <sup>1</sup>	Mean	SEM	
Milk yield, kg d <sup>-1</sup>	34.6	1.0	33.4	1.0	0.12
Fat, g kg <sup>-1</sup> DM	36.7	1.2	35.3	1.1	0.09
Protein, g kg <sup>-1</sup> DM	31.8	0.6	31.6	0.6	0.65
SNF <sup>2</sup> , g kg <sup>-1</sup> DM	89.1	0.7	88.5	0.7	0.18
Milk urea, mg N dl <sup>-1</sup>	16.4	0.3	13.4	0.3	< 0.001

<sup>1</sup>SEM = standard error of the mean.

<sup>2</sup>Non-fat solids.

## Conclusions

Dairy cow performance on reed canarygrass-kura clover mixtures, in grazing or silage feeding systems, is similar to performance on standard pastures or alfalfa silage in Wisconsin. The fields from which data were collected for this research have now been grazed or harvested for silage for 10 years and stands of clover and reed canarygrass remain excellent. This mixture could be a truly permanent component of forage-livestock production systems in northern environments.

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# Evaluation of galega suitability for cattle feeding

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## Abstract

The aim of this investigation was to define the suitability of fodder galega (*Galega orientalis* Lam.) for feeding to different groups of livestock. Due to its longevity, high productivity and good feed value, fodder galega is propagated successfully in Lithuania, Estonia, and Latvia and in other European and American countries. It is therefore important to evaluate its edibility by various cattle and its affect on increasing daily liveweight gain. Experiments to determine galega suitability for stall-feeding and grazing were carried out at the Lithuanian University of Agriculture Training Farm under farm conditions in 1999–2001, and plant chemical analyses carried out. Galega fresh mass and hay were suitable for different livestock feeding because of good edibility. Galega increased daily liveweight gains of heifers under free-grazing (to 705 g) and under stall-feeding (to 873 g) and similarly for steers (to 779 g and 892 g respectively), gains that were typically 68–78 g more than control groups fed on other cultivated grassland species.

Keywords: galega, feed value, edibility, livestock

## Introduction

The legume fodder galega (*Galega orientalis* Lam.) was described in 1788 and is now included in the ISTA List of Stabilized Plant Names (1988), ISTA International Rules of Seed Testing (1996) and OECD List of Varieties (2004). It is a valuable perennial and productive crop with good chemical composition is a very promising crop in Lithuania and in countries with similar geo-climatic conditions (Nõmmsalu, 1994; Balezientiene and Mikulioniene, 2006). Since the 1980s fodder galega has been used on farms in several countries due to its high productivity, protein content and longevity. It can persist for 15 or more years and produces 60–80 t ha<sup>-1</sup> of fresh mass (FM) or 12–20 t ha<sup>-1</sup> of dry matter (DM) based on long-term field trials at the Research Station of Lithuanian University of Agriculture. Forages made of fodder galega contain 14–26% of protein depending on phenologic stages (Szyszkowska et al., 2004). Skórko–Sajko et al. (2005) report that the contents of PDIA, PDIN and PDIE in galega forage were slightly higher than in lucerne forage and silage, but the energy values were similar. Galega FM contains substantial amounts of biologically active materials: vitamins, especially ascorbic acid and carotenes, and minerals. Raig (1994) reports that galega contains substances (alkaloid galegin, etc.) that stimulate lactation by ca. 13%. The increase in milk fat by 0.2–0.3%, and of daily liveweight gain of pigs by an average of 11%, was due to galega's high feed value (Liekus, 1996). The aim of this investigation is to research the chemical composition and edibility of galega FM, and to compare these indices with soya; to define suitability of fodder galega for feeding to different livestock groups and its impact on daily liveweight gain.

## Materials and methods

For the purpose of enriching livestock rations with protein the edibility and feeding of galega were investigated. The FM was composed not only of pure galega, but other grasses and weeds were also present in the tested forages.

Chemical composition of galega DM was determined according to the commonly used Wende forage analyses in the Tempus Laboratory of the Lithuanian University of Agriculture (LUA). Content of amino acids was determined by chromatography, in accordance with EG-Dokument 2354/VI/82, 84/4/EWG and 92/84/EWG. Feeding (cattle and pigs) and grazing (heifers, steers) trials were carried out at the LUA Training Farm and at farms in Sakiai and Marijampole. Investigation of galega FM and hay edibility were carried out with cattle, pigs and dairy cows in stockyards. Edibility was evaluated using a 5-point system by weighing the share of unconsumed fodder: 5 points – very good (80-100%), 4 – good (60-80%), 3 – adequate (40-60%), 2 – poor (20-40%), 1 – bad, only occasionally (5-20%) and 0 – no edibility of grass. According to the age and weight, 10 steers and 20 heifers were evaluated for FM feeding and grazing. The different groups of Lithuanian black and white cattle were tested. Control groups (C) of steers and heifers were fed with grass of pastures (other cultivated species) during 20 and 26 days respectively, and experimental groups (E) – with galega–grass mixture (1:1). Two groups of 12 heifers and steers were compared in grazing experiments. The control groups of cattle were grazed for 45 and 40 days in cultural (sown) pasture, and the experimental groups in pasture where galega content was 50-55%, timothy was 15%, and fescue and other grasses also 15%. Significance ( $P < 0.05$ ) of the data was calculated by dispersion analysis using ANOVA.

## Results and discussion

The chemical analysis data show a high protein content, on average  $231.0 \text{ g kg}^{-1}$  DM, in galega FM. Content and quantity of amino acids, especially essential amino acids, is important for forage quality. The highest amino acid concentrations were for asparagine, glutamine and phenylalanine. In galega these acids were found to constitute 56.37-68.22% of their concentrations in soybean cake; therefore 1 kg soybean cake could substitute for the 1.47–1.72 kg of galega FM (Table 1). Galega DM could substitute for 1 kg soybean cake by equivalent amounts according to the following amino acid concentrations: histidine 1.5 kg, phenylalanine 1.8 kg, threonine 2.5 kg, tyrosine and leucine 2.6 kg, valine 2.7 kg and lysine 2.8 kg. The high quantities of amino acids in the fresh mass of fodder galega indicated that this crop could be used to increase protein content of livestock rations, and could successfully replace soybean cake, which is imported and expensive.

Table 1. Content of amino acids in dry matter of fodder galega.

Amino acid	Content, $\text{mg kg}^{-1}$	Compared with soya, %	Equivalent of 1 kg soya cake, kg	Sequence by amino acid content among 72 fodders
Arginine	6.58	22.38	4.468	23
Asparagine	18.83	68.22	1.466	24
Phenylalanine	10.09	56.37	1.724	17
Glutamine	15.06	33.69	2.968	28
Histidine	6.66	65.29	1.532	21
Isoleucine	4.42	23.02	4.344	21
Leucine	6.36	38.31	2.610	16
Lysine	9.62	36.37	2.767	17
Methionine	3.56	62.46	1.601	9
Serine	7.69	35.93	2.783	19
Tyrosine	6.36	38.31	2.610	16
Threonine	6.67	40.18	2.489	19
Valine	6.58	37.18	2.690	21
LSD <sub>05</sub>	1.02	0.74	0.61	

The assay showed that galega is highly edible, particularly galega FM (60-80%; 4 points). Galega hay was less edible by dairy cows because its dried stems are bulky and rigid. The feeding trial data showed that galega is suitable for both cattle and pigs. Free-grazing and



stall-feeding with galega significantly increased heifer daily liveweight gains (LWG) to 705 and 873 g, respectively, compared with the control (Tables 2 and 3). Heifer daily LWG was increased by 74 and 71 g in comparison with grass pasture and grass stall-feeding (control).

Table 2. Cattle liveweight gains at stall-feeding with galega FM.

	Before feeding, kg	Before feeding, kg	After feeding, kg	After feeding, kg	Daily gain, g	Daily gain, g	Compared with control, g
Cattle	C	E	C	E	C	E	
Steer	325	320	356	399	814	892	78
Heifer	315	310	349	395	802	873	71
LSD <sub>05</sub>	10.6	10.4	27.3	14.2	34.7	30.6	

C – control group; E – experimental group.

The daily LWG of steers fed galega, in stall-fed and grazed trials, were on average 892 and 779 g, respectively, and they exceeded by 78 and 68 g respectively, the LWG of steers on the cultural grass feeding (control). The better feed value, especially the high protein concentration of galega, increased daily LWG of both cattle groups.

Table 3. Cattle liveweight gains under grazing of galega.

	Before grazing, kg	Before grazing, kg	After grazing, kg	After grazing, kg	Daily gain, g	Daily gain, g	Compared with control, g
Cattle	C	E	C	E	C	E	
Steer	243	238	259	287	711	779	68
Heifer	228	212	268	279	631	705	74
LSD <sub>05</sub>	11.4	11.4	14.2	14.2	30.6	30.6	

C – control group; E – experimental group.

## Conclusions

The quantities of amino acids in galega could enrich the protein concentration of livestock rations and replace expensive soybean cake. Protein-rich chemical composition supports favourable edibility of different kinds of forages made from galega. There was good edibility (60-80% consumed) of fodder galega fresh material (FM) by all groups of livestock. Feeding method had no significant influence on daily liveweight gains of heifers and steers, but forages did. Galega FM significantly increased cattle daily gains. Free-grazing and stall-feeding with galega increased heifers daily liveweight gains by 74 and 71 g respectively, and for steers by 68 and 78 g, in comparison with the control (with intake of cultural pasture grass).

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# The effects of forage type and maturity on digesta kinetics of large and small particles in dairy cows

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## Abstract

Digesta kinetics of large and small particles were estimated in steady-state conditions using five dairy cows in a 5 × 5 Latin square experiment. Two grass (timothy-meadow fescue) and two red clover silages harvested at early and late growth stages and a mixture of late grass and early red clover were used as dietary treatments. Cows received the silages *ad lib* and 9.0 kg concentrate per day. Ruminal contents were evacuated two times in each experimental period. The total faecal collection was performed during d 18-21 of each period. Ruminal and faecal particulate matter was divided into large (> 1.25 mm) and small (1.25-0.038 mm) particles by wet sieving technique. Indigestible NDF (iNDF) was determined by a 12d ruminal *in situ* incubation (pore size of 0.017 mm) followed by NDF extraction. Forage type and maturity did not markedly affect ruminal particle size distribution. Pool size of iNDF in the rumen was greater with red clover compared to grass silage diets. Passage rates of iNDF and potentially digestible NDF (pdNDF) increased with decreasing particle size. Passage rates of iNDF and pdNDF in small particles and particle breakdown rate were slower in red clover compared with grass silage diets, but digestion rate of pdNDF was faster. Contribution of particle breakdown to turnover of different particle size fractions of rumen iNDF was lower in red clover compared with grass silage diets and it decreased with advancing forage maturity. Mixing the silages did not change the ruminal iNDF and pdNDF contents and their ruminal kinetics compared to the average of pure silages.

Keywords: steady-state model, indigestible NDF, passage rate, breakdown rate, legume, grass

## Introduction

The rates of digestion and passage are important determinants of fibre digestibility. *In situ* and *in vitro* incubations have usually been used to estimate the rate of digestion ( $k_d$ ), while rate of passage ( $k_p$ ) is usually estimated using marker techniques. These estimated rate constants should be compared with an integrated rumen model such as rumen evacuation technique, because  $k_d$  estimations derived from *in situ* incubation have been lower than those derived from rumen evacuation technique (Bruining *et al.*, 1998).

Neutral detergent fibre (NDF) is not a homogenous entity because indigestible NDF (iNDF) and potentially digestible NDF (pdNDF) have different kinetic characteristics, e.g. pdNDF is selectively retained in the rumen (Huhtanen *et al.* 2006). Rumen evacuation technique together with wet sieving technique and iNDF determination can be used to study  $k_p$  of iNDF and  $k_p$  and  $k_d$  of pdNDF in both escapable and non-escapable ruminal pools (Huhtanen *et al.*, 2007).

Grasses and legumes differ in digesta kinetics due to different histological characteristics, and forage maturity also affects them. The purpose of this study was to compare ruminal pool sizes and digesta kinetics of escapable and non-escapable particles in dairy cows fed grass and red clover silage diets harvested at two contrasting stages of growth.



## Materials and methods

Five ruminally fistulated multiparous Ayrshire dairy cows with the milk production of  $27.1 \pm 0.9 \text{ kg d}^{-1}$  were assigned randomly to a  $5 \times 5$  Latin square design with 21 d periods. Experimental silages were prepared from primary growths of timothy-meadow fescue and red clover swards harvested at early or late stages of growth in Jokioinen, Finland in 2003. The grass silages were harvested on 17 June at early ( $G_E$ ) and on 26 June at late ( $G_L$ ) growth stages and the red clover silages were harvested on 2 July at early ( $R_E$ ) and on 16 July at late ( $R_L$ ) growth stages. The forages were fed *ad libitum* and supplemented with  $9.0 \text{ kg d}^{-1}$  of barley and oats-based concentrate. Diets were offered four times daily. Ruminal contents were evacuated at 6 and 12 h on d 13 and 15 of each experimental period, respectively. The average weight of ruminal contents of the two evacuations was used as the estimation of the diurnal mean. The total faecal collection was performed during d 18-21 of each period.

Particle size distribution of ingested silages was adopted from Rinne *et al.* (2002). The particle size distribution of dietary concentrate, ruminal digesta and faeces was determined by a wet sieving apparatus (Retsch AS200 Digit, GmbH, Haan, Germany). The samples were divided into large (LP,  $>1.25 \text{ mm}$ ) and small (SP,  $1.25\text{--}0.038 \text{ mm}$ ) particles by wet sieving. After drying the materials were ground ( $1 \text{ mm}$ ). The NDF concentration was analysed by ANKOM<sup>220</sup> Fiber Analyser. To determine the iNDF concentration,  $1.0 \text{ g}$  of sample was weighed into nylon bags ( $60 \times 120 \text{ mm}$ , pore size of  $0.017 \text{ mm}$ ) and duplicate bags were incubated for 12 d in the rumen of two cows fed a forage based diet. The pdNDF concentration ( $\text{g kg}^{-1} \text{ DM}$ ) was calculated as  $\text{NDF} - \text{iNDF}$ . Calculations of ruminal pool sizes and kinetics of iNDF and pdNDF are described by Huhtanen *et al.* (2007). No NDF was assumed in the material  $< 0.038 \text{ mm}$ . The Latin square split-plot model was used to analyse the data (PROC Mixed; SAS, 2003), where the treatment and particle size were considered as the main and sub plots, respectively. Orthogonal contrasts were used to compare the diets.

## Results and discussion

The NDF and iNDF concentrations in  $G_E$ ,  $G_L$ ,  $R_E$ ,  $R_L$  and the dietary concentrate were 500, 570, 375, 463, 205, and 57, 84, 70, 138,  $70 \text{ g kg}^{-1} \text{ DM}$ , respectively. The amount of ruminal particulate DM was similar in LP ( $4.70 \text{ kg}$ ) and SP ( $4.69 \text{ kg}$ ) (Table 1). It was greater with grass ( $4.93 \text{ kg}$ ) compared to red clover ( $4.32 \text{ kg}$ ) silage diets and increased with advancing maturity. However, ruminal iNDF content was greater with red clover ( $1.95 \text{ kg}$ ) compared to grass silage diets ( $1.14 \text{ kg}$ ). It increased with advancing maturity and the maturity effect was greater with red clover compared to grass silage diets. The changes reflect the general trends of iNDF (and lignin concentration), which is higher in legumes and increases with increasing maturity of plants. The content of pdNDF in ruminal LP was greater than that in SP ( $2.35$  vs.  $1.77 \text{ kg}$ ) and the difference was greater with grass compared to red clover silage diets. Mixing the silages tended ( $P = 0.06$ ) to increase the ruminal particulate DM content compared to the pure silages. Mixing the silages did not affect ruminal iNDF and pdNDF contents.

The  $k_p$  of iNDF and pdNDF was faster for grass ( $0.023$  and  $0.012$ , respectively) compared to red clover ( $0.020$  and  $0.006$ , respectively) silage diets and for SP ( $0.038$  and  $0.016$ ) compared to LP ( $0.005$  and  $0.003$ ). LP must be broken to a specific size to be eligible for passage through the reticulo-omasal orifice. The  $k_r$  was greater for grass compared to red clover silage diets ( $0.030$  vs.  $0.018$ ) and increased with advancing maturity ( $0.021$  vs.  $0.027$ ). The contribution of particle breakdown in clearance of iNDF from ruminal LP, calculated as  $k_r(k_r + k_p)^{-1}$  of iNDF, was greater with grass compared to red clover silage diets ( $0.85$  vs.  $0.78$ ).

The  $k_d$  of pdNDF for red clover was faster than that for grass silage diets ( $0.081$  vs.  $0.040$ ) and it decreased with mixing of the silages compared to the average of the pure silages. The faster  $k_d$  of pdNDF for red clover compared to grass silage diets is consistent with Bruining *et al.*

(1998). The contribution of digestion in clearance of pdNDF from ruminal pools, calculated as  $k_d (k_r + k_p + k_d)^{-1}$ , was greater for red clover compared to grass silage diets, increased with decreasing particle size and increased with mixing the silages. It can be concluded that escapable particles have different kinetic parameters in the rumen. Different forage types should be taken into account in the models predicting feed intake and fibre digestibility.

Table 1. Ruminal pools and faecal excreted amounts of dry matter (DM), indigestible NDF (iNDF) and potentially digestible NDF (pdNDF), and ruminal rates of intake ( $k_i$ ), passage ( $k_p$ ), particle breakdown ( $k_r$ ) and digestion ( $k_d$ ) of large (LP, >1.25 mm) and small (SP, 1.25-0.038 mm) particles of dairy cows fed diets based on different silages.

	Grass silage						Red clover silage						Orthogonal contrasts <sup>2</sup>					
	Early		Late (G <sub>L</sub> )		G <sub>L</sub> R <sub>E</sub>		Early (R <sub>E</sub> )		Late		SEM <sup>1</sup>							
	LP	SP	LP	SP	LP	SP	LP	SP	LP	SP								
Ruminal digesta, kg																		
DM	4.65	4.75	5.05	5.26	5.15	4.84	4.00	3.56	4.66	5.05	0.306	**	**	ns	†	ns		
iNDF	0.88	1.24	0.99	1.44	1.40	1.63	1.64	1.40	2.36	2.41	0.117	**	**	**	ns	**		
pdNDF	2.93	2.27	3.17	2.41	2.71	1.84	1.49	1.09	1.46	1.24	0.148	**	ns	ns	ns	**		
Faeces, kg d <sup>-1</sup>																		
DM	0.35	2.90	0.48	3.57	0.42	2.93	0.32	1.86	0.41	2.89	0.144	**	**	ns	ns	**		
iNDF	0.11	1.17	0.14	1.40	0.12	1.46	0.18	1.13	0.28	1.96	0.051	**	**	**	†	**		
pdNDF	0.19	1.03	0.26	1.35	0.20	0.71	0.08	0.27	0.07	0.28	0.101	**	ns	ns	ns	**		
iNDF kinetics																		
<i>k<sub>i</sub></i> , h <sup>-1</sup>	0.034	0.020	0.039	0.019	0.030	0.017	0.019	0.018	0.026	0.014	0.0021	**	ns	ns	ns	**		
<i>k<sub>p</sub></i> , h <sup>-1</sup>	0.005	0.042	0.006	0.041	0.003	0.039	0.005	0.034	0.005	0.035	0.0023	**	ns	ns	ns	**		
<i>k<sub>r</sub></i> , h <sup>-1</sup>	0.028		0.032		0.025		0.014		0.021		0.0018	**	**	ns	ns	-		
<i>k<sub>r</sub></i> ( <i>k<sub>r</sub></i> + <i>k<sub>p</sub></i> ) <sup>-1</sup>	0.85		0.84		0.86		0.75		0.81		0.016	**	ns	†	*	-		
pdNDF kinetics																		
<i>k<sub>i</sub></i> , h <sup>-1</sup>	0.075	0.032	0.069	0.030	0.076	0.038	0.091	0.048	0.104	0.047	0.0040	**	ns	ns	ns	**		
<i>k<sub>p</sub></i> , h <sup>-1</sup>	0.003	0.020	0.003	0.023	0.004	0.017	0.002	0.011	0.002	0.010	0.0022	**	ns	ns	ns	**		
<i>k<sub>d</sub></i> , h <sup>-1</sup>	0.044	0.037	0.038	0.039	0.039	0.045	0.077	0.089	0.084	0.074	0.0059	**	ns	ns	**	ns		
<i>k<sub>d</sub></i> ( <i>k<sub>r</sub></i> + <i>k<sub>p</sub></i> + <i>k<sub>d</sub></i> ) <sup>-1</sup>	0.59	0.65	0.52	0.63	0.57	0.72	0.82	0.89	0.79	0.88	0.033	**	ns	ns	*	**		

<sup>1</sup> SEM, standard error of means, n = 50 ns, non significant; †,  $P < 0.10$ ; \*,  $P < 0.05$ ; \*\*,  $P < 0.01$ .

<sup>2</sup> C<sub>1</sub> Grass vs. red clover, C<sub>2</sub> Early vs. late harvest time, C<sub>3</sub> Interaction of forage species and forage maturity, C<sub>4</sub> Mixture of G<sub>L</sub> and R<sub>E</sub> vs. G<sub>L</sub> and R<sub>E</sub> fed separately, C<sub>5</sub> Large vs. small particles.

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# Effects of chop length of silage on chewing time, feed consumption and milk production

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## Abstract

Grass silage of high quality is a natural prerequisite for modern European dairy production. Physical structure of silage is important for animal performance as well as for animal welfare. Allowing a longer cutting length of the silage decreases the energy used for harvesting the silage. This must, however, not be at the expense of feed intake or milk production.

Grass silage of high nutritional quality (11.5 MJ ME kg<sup>-1</sup> DM; 157 g CP g kg<sup>-1</sup> DM; ~400 g kg<sup>-1</sup> DM) was harvested with two different types of self-loading wagons giving different physical structures. A mechanical separation showed relatively small differences in half weight length (34 or 25 mm) but the upper quartile differed more (57 vs. 39 mm). Image analyses for the same silage showed mean lengths of 57 and 35 mm.

The silage was fed to dairy cows of different parity and stages of lactation. Pattern of consumption and chewing time was measured using IGER animal behaviour recorders. The most obvious difference between the two silages was longer chewing time per kg DM silage for the longer silage (51 vs. 43 minutes). However, the average daily silage consumption did not differ significantly between treatments (13.6 vs. 13.9 kg DM cow<sup>-1</sup>) and neither did the total feed intake (24.0 vs. 24.4 kg DM). Overall daily milk production (kg ECM cow<sup>-1</sup>) was similar for the treatments (31.5 vs. 30.8). The conclusion from the present experiment is that, given high quality silage, differences in physical structure within the limits of the experiment do not restrict feed intake and milk production.

Keywords: Self-loading wagon, physical structure, feed intake, dairy cows

## Introduction

Modern high-yielding dairy cows have high demands for feed in order to produce and function optimally. Silage of high nutritional and hygienic quality is a necessity. Technical developments in silage techniques have again placed focus on the length of the chopped silage. Earlier Swedish research emphasized the lower limit for chop length and the general recommendation was >20 mm nominal chop length to ensure enough physical structure in the ration (Lindell *et al.*, 1970). Today energy consumption in agriculture is a key issue. Of course, less fuel energy is required for the coarser structure of silage. This must, however, not be at the expense of feed intake and animal production. The aim of the present trial was to study the effects of chop length of grass silage and possible interaction with fibre source in concentrate. Only parts of the total study are reported in the present paper.

## Materials and methods

A first cut of a grass dominated sward (timothy, meadow fescue) was harvested on June 8-9 2005 at Kungsängen Research Centre, Uppsala, Sweden. Two self-loading wagons, either a Pöttinger Jumbo 7200 L rotor cutter (P wagon) or a Taarup/Sahlström precision chopping wagon (T wagon) was used. Technical data and ensiling results were also studied and are reported in this volume by Knicky and Lingvall (2007).

Physical structure was determined on the silages as fed (samples taken at feeding), i.e. after the silage passed through a mechanical unloader in the silo and after been distributed through a feeding wagon. Three methods were used: 1) a Penn State Separator (Heinrichs and Kononoff, 2002); 2) screening of dry samples (Gale and O'Dogerty, 1982); 3) image analyses (Nørgaard, 2006). The two silages were fed to 56 dairy cows of the Swedish Red Breed equally distributed to the two silages for up to 20 weeks (average ~16 weeks). Cows from different lactation stages were included in the trial. The cows were in a barn equipped with an automatic milking unit, and milk was registered automatically at all milkings, as were feeds (Spörndly and Wredle, 2004). Silage and concentrate were fed separately and amounts of concentrate were based on milk production and silage intake before the start of the experiment. Chewing time and pattern were monitored for 18 cows during one 24-hour period using an IGER animal behaviour recorder (Ultra Sound Advice, London, UK). Statistical analyses were carried out for this part of the study with the mixed procedure of SAS (SAS Institute, Raleigh, N.C., USA). For the rest of the study only general means are presented.

## Results and discussion

Some figures on chemical composition, calculated content of metabolizable energy and silage pH are presented in Table 1. Further figures on silage quality are presented by Knicky and Lingvall (2008). The silages were of exceptionally high nutritional quality and DM content was optimal for obtaining high silage intake. NDF content in the silage was similar for both silages, ~450 g kg<sup>-1</sup> DM.

Table 1. Chemical composition, calculated content of metabolizable energy (ME) and silage pH (n = 9). Mean (std. dev)

	P wagon (rotor cutter)	T wagon (precision chopping)
DM, g kg <sup>-1</sup> DM	389 (20)	404 (20)
Crude protein, g kg <sup>-1</sup> DM	157 (8)	154 (6)
Ash, g kg <sup>-1</sup> DM	93 (3)	93 (7)
Metabolizable energy, MJ ME kg <sup>-1</sup> DM	11.5 (0.1)	11.5 (0.1)
pH	4.4 (0.1)	4.4 (0.1)
Ammonia - N, g kg <sup>-1</sup> N	48 (2)	45 (4)

The Penn State Separator was not suited for this sort of material. On average, more than 90% of the silage was found on the upper screen. Results from the separation according to Gale and O'Dogerty's (1982) screen procedure showed that half weight lengths were on average (n = 3) 34 and 25 mm; upper quartile, 57 and 39 mm; and, lower quartile 20 and 16 mm for the P and T wagons, respectively. The image analysis showed median values of 56 and 35 mm and arithmetic means of 57 and 35 mm for the P and T wagons, respectively. These figures represent the silages as fed and are of course influenced by handling of the feeds after ensiling, which is unavoidable in practical feeding. There is, however, no reason to believe that the handling of silages should affect the silages different, i.e. that the basic difference is due to the differences in harvesting technique.

Results from the chewing measurements (n = 18) are shown in Table 2. The most obvious result was that chewing time for silage from wagon P was higher than for wagon T ( $P < 0.06$ ). It must of course be kept in mind that these measurements represent only a small part of the total study and it is therefore not surprising that feed intake diverge from that for the whole study.

Results from the feeding trial (n = 56) are presented in Table 3. Only overall means are included (no statistical measurements). As could be observed, the results are very similar for the two treatments. In the final statistical analyses, the different lengths of the experimental

period for individual cows and interaction between silage and concentrate intake will be accounted for. Considering the large number of cows, the overall trend will probably not change.

Table 2. Feed consumption (kg DM), daily chewing time and mean chewing time and *P*-values. LS-means (se) for one 24-hour period (Bergfors, 2006).

	P Wagon (rotor cutter)	T Wagon precision cutting)	<i>P</i> <
Silage intake, kg DM d <sup>-1</sup>	15.3 (0.9)	17.1 (0.8)	0.15
Total intake, kg DM d <sup>-1</sup>	24.1 (1.9)	27.5 (1.7)	0.21
Daily chewing time, min. d <sup>-1</sup>	810 (36)	786 (31)	0.62
Daily chewing time (concentrate deducted), min. d <sup>-1</sup>	765 (35)	733 (30)	0.49
Mean chewing time min. kg <sup>-1</sup> total DM intake	35 (3)	30 (2)	0.16
Mean chewing time, min. kg <sup>-1</sup> silage DM intake	51 (3)	43 (3)	0.06

Table 3. Feed intake and milk production when feeding silages of different chop lengths (P vs. T). Means (standard deviation) for the whole experimental period.

	P wagon (rotor cutter)	T wagon (precision cutting)
<u>Feed intake</u>		
Silage intake, kg DM d <sup>-1</sup>	13.6 (2.0)	13.9 (4.0)
Concentrate intake, kg	10.4 (3.7)	10.5 (4.0)
<u>Milk yield</u>		
Milk, kg d <sup>-1</sup>	29.7 (8.7)	28.8 (7.2)
ECM, kg d <sup>-1</sup>	31.5 (7.5)	30.8 (7.0)
Fat, g kg milk <sup>-1</sup>	46 (6)	46 (5)
Protein, g kg milk <sup>-1</sup>	36 (4)	36 (3)
Lactose, g kg milk <sup>-1</sup>	46 (1)	46 (1)

## Conclusions

The difference in chop length between these two silages meant that cows needed longer time to chew and consume the silage. Given the high nutritional value of the silage this was no limitation for feed intake and milk production.

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# Performance of continental weanling heifers grazing pasture or consuming grass silage during winter in Ireland

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## Abstract

Liveweight gain (LWG) and dry matter (DM) intake of continental weanling heifers grazing pasture or consuming grass silage in winter were examined in Ireland. Fifty spring-born, Charolais × Limousin weanlings with an initial mean live weight (LW) of 275 kg were used. For 141 d, 25 heifers grazed a pasture allowance of ~3.5 kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup> and 25 heifers were housed and offered grass silage *ad libitum*. Mean LWG of the grazed heifers was 0.14 kg d<sup>-1</sup> higher (0.59 vs. 0.45 kg d<sup>-1</sup>) and consequently their LW at the end of winter was 25 kg greater (358 vs. 333 kg) than those on grass silage. Mean apparent DM intake of the grazed heifers was ~1 kg DM heifer<sup>-1</sup> d<sup>-1</sup> higher (6.3 vs. 5.2 kg heifer<sup>-1</sup> d<sup>-1</sup>). The differences in LWG and apparent DM intake resulted in a mean feed conversion efficiency of 0.095 and 0.086 kg LWG kg DM intake<sup>-1</sup> for pasture and grass silage, respectively. These results suggest that weanling heifers can achieve an acceptable LWG when grazing a pasture allowance of 3.5 kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup> in their first winter of a pasture-based beef system.

Keywords: permanent pasture, winter grazing, grass silage, liveweight gain, feed intake, beef

## Introduction

Perennial pastures form the main source of nutrients for beef cattle in Ireland. However, pasture growth rate varies over the year and in winter insufficient pasture is available to meet the total feed requirements of the breeding herd. Potentially, a method of reducing the requirement for conserved feeds and housing is for weanling cattle to graze in winter pasture which has accumulated since the autumn (Weldon *et al.*, 2006).

During their first winter, a modest target liveweight gain (LWG) (0.4-0.6 kg d<sup>-1</sup>) in weanlings is often desired to minimize feed requirements and exploit compensatory LWG in the following spring. A wide body of information exists about the amount and quality of grass silage that weanlings require to achieve this production target (e.g. Keane, 2002), but there is much less information about their feed requirements when grazing pasture in winter. A good starting point is to estimate the amount of metabolizable energy (ME) the weanlings require using published data for grazing livestock (Nicol and Brookes, 2007), in order to estimate their daily pasture allowance and plan their winter grazing management.

Thus, the initial objective of this study was to estimate the ME and pasture allowance required for a continental weanling heifer to grow at 0.5 kg d<sup>-1</sup> when grazing pasture in winter. The second objective was to compare the performance of weanling heifers grazing the estimated pasture allowance with those housed and offered grass silage *ad libitum* in winter.

## Materials and methods

The experiment was conducted at the Teagasc Beef Research Centre, Grange, County Meath, Ireland (53°30'N, 6°40'W, 92 m above sea level). Fifty weaned, spring-born Charolais × Limousin heifers were sourced from Irish farms through a livestock mart. All heifers were treated for diseases and parasites. Initial mean (SD) live weight (LW) was 275 (27.0) kg and the mean age was 252 (28.0) d. On 28 November, heifers were stratified into 25 weight blocks

(light to heavy). Two treatments of pasture and grass silage were assigned randomly to heifers within each block. The duration of the study was 141 d from 28 November 2006 to 18 April. The ME requirement for the heifers growing at the target LWG of  $0.5 \text{ kg d}^{-1}$  was estimated using tabulated data (Nicol and Brookes, 2007) derived from the results of numerous experiments published in authoritative publications, e.g. those from the United Kingdom (AFRC, 1990). While these estimates are based on a range of assumptions and are subject to inherent between-animal variation, the values given can be considered a good starting point. For the pasture treatment, heifers were rotationally stocked in three replicate groups (eight or nine heifers group<sup>-1</sup> balanced for initial LW) on pasture cover accumulated since the autumn (mainly *Lolium perenne* L., *Poa* spp. and *Trifolium repens* L.). Using portable electric fences to sub-divide existing fields, the heifers were offered the fresh pasture allowance daily and did not have access to the previous day's pasture. Pasture allowance and a post-grazing pasture mass of 800-900 kg DM ha<sup>-1</sup> were achieved by varying the size of the area grazed each day. Drinking water was supplied in portable troughs. For the silage treatment, heifers were housed in five available pens (five heifers pen<sup>-1</sup> of 11 m<sup>2</sup>) in a slatted-floor shed. Heifers were grouped into five weight blocks and each group was assigned at random to a pen. Grass silage was offered daily *ad libitum*. Each pen was supplied with drinking water. All heifers were weighed at 19-35 d intervals and LWG was calculated for each interval and the overall period. For each replicate group, apparent dry matter (DM) intake was estimated as the difference between mass (kg DM) of available and residual feed (after 24 h) divided by the number of heifers per group. Pasture mass was estimated using a rising plate meter (Jenquip Ltd., Fielding, New Zealand) calibrated on five dates during the study. Associations between plate meter readings and pasture mass (measured to ground level) were strong ( $0.94 < R^2 < 0.99$ ) and highly significant ( $P < 0.001$ ) for each calibration date. Feed conversion efficiency (FCE) was calculated from mean LWG and apparent DM intake for each group. Data were analysed using a mixed effects model:  $Y_{ij} = \mu + T_i + b_j + \varepsilon_{ij}$ , where  $Y_{ij}$  is the response variable of an individual heifer (LW, LWG) or group (DM allowance, DM intake, FCE) with the  $i$ th treatment in the  $j$ th weight block or group,  $\mu$  is the overall mean,  $T_i$  is the fixed effect of treatment,  $b_j$  is the random effect of block or group, and  $\varepsilon_{ij}$  is the error terms.

## Results and discussion

Based on published ME requirements (Nicol and Brookes, 2007), we estimated that a late maturing heifer with a mean LW of 300 kg growing at  $0.5 \text{ kg d}^{-1}$  would require 64 MJ ME d<sup>-1</sup> or 5.8 kg DM d<sup>-1</sup> of pasture with an ME concentration of 11 MJ ME kg DM<sup>-1</sup>. Assuming 0.5-0.6 of the pre-grazing pasture mass is utilised during grazing in winter, the heifer would require a pasture allowance of approximately 3.0-4.0 kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup>. Because actual requirements were likely to differ among animals, a target pasture allowance of 3.5 kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup> was used to preclude the possibility of greatly underfeeding some animals. The actual DM allowance increased over time (Table 1), which satisfied our criteria for a pasture allowance of ~3.5 kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup> and *ad libitum* grass silage. The mean LWG of the grazed heifers was 0.14 kg d<sup>-1</sup> higher ( $P < 0.01$ ) and consequently their LW at the end of winter was 25 kg greater ( $P < 0.001$ ) than those on grass silage. However, the mean apparent DM intake of the grazed heifers was also about 1 kg DM heifer<sup>-1</sup> d<sup>-1</sup> higher ( $P < 0.001$ ), which resulted in a similar ( $P = 0.20$ ) mean FCE for the two diets. The mean LWG of 0.45 kg d<sup>-1</sup> for the silage treatment was better than results reported previously (0.2-0.4 kg d<sup>-1</sup>) for similar weanlings housed and offered grass silage without concentrates (Keane, 2002). There was an interaction ( $P < 0.001$ ) between the feeds and sampling periods in their effects on LW and LWG (Table 1). The LW of heifers on both feeds increased over time, but LWG was particularly higher ( $P < 0.01$ ) for heifers on pasture in the fourth and fifth periods. This result was associated with increases in apparent DM intake over time for both feeds. While



there were differences ( $P < 0.05$ ) in FCE between feeds in the second and fifth periods, it is difficult to establish the causes of these results. Further analyses of the nutritive value of the two feeds are needed to determine any impacts of feed quality.

The implication for beef systems is that weanling heifers can achieve acceptable weight gains when grazing a pasture allowance of  $\sim 3.5$  kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup> in their first winter. An autumn pasture budget which incorporates estimates of the ME or feed demands of the grazed weanlings, and pasture supply, should be recommended to determine the pasture allowance and to establish a winter grazing management plan. Pasture supply should be carefully rationed to increase the length of the grazing rotation and therefore allow sufficient time for grazed pasture to recover before it is grazed again in early spring. The capacity of a beef farm to utilise 'out-of-season' pasture is likely to depend on autumn and winter pasture supply, soil and climate conditions, and the feed requirements of different classes of livestock in the herd.

Table 1. Mean dry matter (DM) allowance, final live weight (LW), liveweight gain (LWG), apparent DM intake and feed conversion efficiency (FCE) of continental weanling heifers grazing pasture or consuming grass silage during winter (141 d) in County Meath, Ireland.

Variable	Treatment	28 Nov.- 2 Jan.	3 Jan.- 22 Jan.	23 Jan.- 19 Feb.	20 Feb.- 22 Mar.	23 Mar.- 18 Apr.	Mean (141 d)
DM allowance (kg d <sup>-1</sup> )	Pasture	8.3	9.9	11.0	11.3	12.6	10.9
	Silage	4.9	5.2	5.8	5.8	6.7	5.8
	SED	0.11	0.24	0.22	0.21	0.25	0.13
	<i>P</i> value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Final LW (kg)	Pasture	292	294	308	339	358	311
	Silage	281	291	306	326	333	302
	SED	3.3	3.0	4.4	5.7	6.5	1.6
	<i>P</i> value	0.003	0.362	0.601	0.027	0.001	< 0.001
LWG (kg d <sup>-1</sup> )	Pasture	0.47	0.12	0.51	1.00	0.69	0.59
	Silage	0.14	0.51	0.52	0.65	0.27	0.45
	SED	0.079	0.107	0.094	0.081	0.070	0.045
	<i>P</i> value	< 0.001	0.001	0.858	< 0.001	< 0.001	0.004
DM intake (kg d <sup>-1</sup> )	Pasture	5.2	6.4	6.9	6.3	6.7	6.3
	Silage	4.3	4.6	5.3	5.3	5.8	5.2
	SED	0.14	0.19	0.15	0.14	0.20	0.08
	<i>P</i> value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
FCE (kg LW kg DM <sup>-1</sup> )	Pasture	0.090	0.016	0.075	0.161	0.102	0.095
	Silage	0.034	0.108	0.099	0.122	0.046	0.086
	SED	0.0254	0.0305	0.0209	0.0217	0.0112	0.0057
	<i>P</i> value	0.070	0.023	0.287	0.120	0.003	0.202

## Conclusion

Continental weanling heifers grazing a pasture allowance of  $\sim 3.5$  kg DM 100 kg LW<sup>-1</sup> d<sup>-1</sup> in winter (141 d) gained more LW than those housed and offered grass silage *ad libitum*, which was associated with a higher apparent DM intake and similar FCE for the grazed heifers.

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# Impacts of compact calvings and once-a-day milking in grassland based systems

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## Abstract

A sustainable production system relies on decreasing the production costs and improving the farmer's working conditions. Two technical solutions were tested during 3 years within a grass based system: compact calvings to close the milking parlour during 1 month, and once-a-day (OAD) milking. A close calving pattern based on a 3-month period appeared to be more repeatable than a 2-month system. Technical and economic results did not significantly differ between spring and autumn calvings. High genetic merit cows can be milked only OAD during the whole lactation without negative impacts on welfare or economic efficiency. Both solutions were very effective in improving work organization.

Keywords: grazing, dairy production, once a day milking, calving period

## Introduction

A sustainable dairy production system relies on decreasing the production costs and improving the farmer's working conditions. One solution consists in implementing low concentrate systems based on an extended grazing season (Brocard *et al.*, 2000). A compact calving pattern should allow the closure of the milking parlour for 1 month. Compact calvings are already widely implemented in countries such as New Zealand but require extra experiments in France (Brocard *et al.*, 2005). Finally, decreasing milking frequency might free farmers from a large part of their daily work and lead to a lower energy deficit in early lactation (Brocard *et al.*, 1998; Pomiès *et al.*, 2002; Dalley *et al.*, 2007).

## Materials and methods

Trevarez experimental unit is located in Brittany in an oceanic climate. The 2002/2005 experimental programme was based on a forage system with 0.5 ha of grazed grass per cow. It included 2 experiments: the first one consisted of comparing autumn calvings (A2 group) to late winter calvings (LW2). A second trial compared the latter with a group milked once-a-day during the whole lactation (LW1 group). In 2002 the Holstein cows were allocated to one group for 3 years. Primiparous cows were introduced yearly to replace culled cows. Each group was initially made of 27 cows including 40% of first lactations. The 151 cows which took part to the experiment over the 3-year period were split into 3 'cohorts': P (only primiparous), PM (primiparous-multiparous) and M (only multiparous) cows. Altogether 80 lactations were completed in the A2 group vs. 80 in the LW2 group and 73 in the LW1 group. The groups were given the same quality and quantity of forages. Rotational grazing was monitored on pure ryegrass or ryegrass-clover associations. Each group was allocated one group of paddocks with the same grazing management rules (sward heights, key-dates, etc). In the A2 group, the average '100% grazed grass' period (no buffer feed) lasted 189 days, the 'day and night' grazing period (buffer feed under 5 kg DM d<sup>-1</sup>) 226 days, and the total grazing period 305 days. In the LW groups, the average '100% grass' period lasted 175 days, the 'day and night' grazing period 200 days, and the total grazing period 288 days. Maize silage was delivered to the lactating cows in winter, while dry cows received a mixed grass and maize

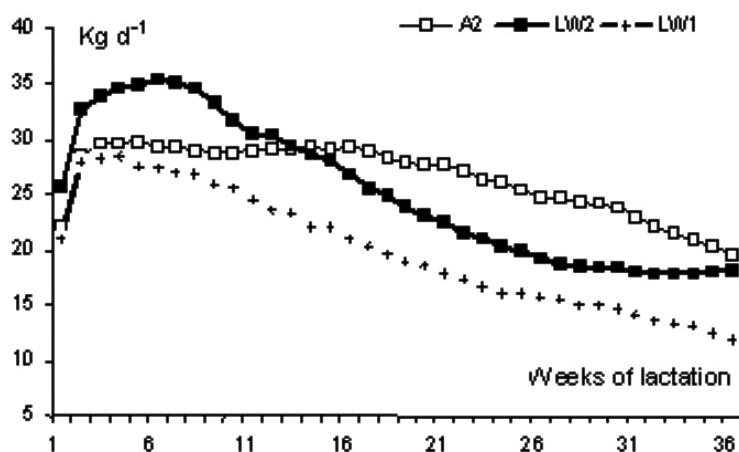
silage diet. Only wheat was used as energy concentrate to simplify diets. The only N concentrate was soyabean in the maize; the N concentration target was 90 g of PDI kg<sup>-1</sup> of DM for lactating cows. The total 'wheat + soya' amount was 2 kg cow<sup>-1</sup> d<sup>-1</sup> from calving to conception. The 3 groups calved in a compact pattern in 2 periods: autumn (average date 20 September) for the A2 group, or late winter (20 February) for the LW groups. The reproduction protocol (Brocard *et al.*, 2005) was based on the exhaustive recording of all events, on diagnoses by the vet before and during the AI periods, and on systematic conception diagnoses. Calvings were grouped without hormones. Cows were dried off according to the closing date of the milking parlour (23 July for A2, 23 December for LW groups) whatever their production. A specific protocol to assess animal welfare (Brulé *et al.*, 2003) was added during 2 years to the usual measurement protocol. The daily work was registered at 4 key dates. Production data were analysed according to a mixed linear model with repeated measures on cows (Brocard *et al.*, 2007). Health troubles and reproductive performances were analysed the following way: binary variables by a logistic model, census variables through an exponential regressive model, and the risk of non conception was analysed by a survival test. The economic analysis was made at the farm gross margin (GM) level and included all the technical results of the experiment. Estimations were made for an average French farm of 280,000 l of quota.

## Results

The yearly intakes of stored forages varied from 2.1 to 2.4 t DM per cow according to the group. As grazing management rules were the same for all the groups, no difference appeared in terms of forage intakes or quality between the 2 calving seasons (Brocard *et al.*, 2005). The average concentrate levels per cow per year in the groups were the following: 465 kg in group A2, 388 in group LW2, and 332 in group LW1. The lactation curves showed very different shapes, autumn calvings leading to a flatter curve (Figure 1). Though at the whole lactation scale multiparous cows of the A2 group produced some 300 extra kg of milk (ns) compared to the LW2 cows. Only the P cows produced significantly more milk (+ 1.6 kg d<sup>-1</sup>,  $P = 0.02$ ) with a higher fat content (+ 2.8 g kg<sup>-1</sup>,  $P = 0.04$ ) when calving in autumn. M and PM cows significantly produced a higher protein content in the A2 group (+ 1.3 g kg<sup>-1</sup>,  $P = 0.04$ ) than in the LW2 group. Compared to the LW2 group, the LW1 cows lost 21% milk per year. All OAD cow cohorts produced significantly less milk per day (-5.5 to -6 kg d<sup>-1</sup>,  $P < 1\%$ ), with higher fat (+ 2.4 to + 3.5 g kg<sup>-1</sup>) and protein (+ 2.3 to + 2.9 g kg<sup>-1</sup>) contents. The average body condition score at calving were close to 3.0 in each group. Losses after calving were close and below 1 pt for all groups. OAD led to a better condition score at the end of the lactation (2.9 vs. 2.6 for group LW2 and 2.5 for group A2). A compact calving pattern within an 8-week period did not appear realistic in any of the 2 groups milked twice daily because of the high culling rate it would lead to: only 58 to 61% of the cows were in calf within 2 months (vs. 81% of the OAD cows). The only significant difference was the shorter days open interval (- 1 d,  $P = 4\%$ ) in the LW1 group. No significant difference was found on the total frequency of health troubles. The LW1 multiparous cows developed significantly more mastitis ( $P = 9\%$ ) but less cases of lameness ( $P = 7\%$ ) than the LW2 group. The culled cows of the LW1 group required a shorter fattening period (-10 d). As they were heavier and had a better conformation their sale price was higher. The main culling reasons were mastitis for the LW1 group and infertility for the LW2 and A2 groups. The yearly working time (WT) reached 1,800 hours for both groups milking twice daily. The compact calving patterns led to a strong monthly organization. Though with 27% extra cows to produce the quota, the OAD system led to a decrease by 17% of the total yearly WT. The LW2 system required 3.7 extra cows and 5.5 extra ha than the A2 system. The farm GM did not significantly differ. With its higher cow numbers (+12 cows) the LW1 group kept low feeding costs (< 45 € per t of milk). The

higher milk price and beef by-products led to a higher dairy GM by 2,730 €. The larger forage area (+ 6.1 ha) decreased the crops area. Hence the farm GM difference was not significant. The potential increase in the investments might be related to the housing of the 12 extra cows: a close spring-calving pattern leads to culled cows in winter, with fewer requirements for cow housing. The higher cow number must also be consistent with the environmental regulations.

Figure 1. Non-adjusted dairy production of multiparous cows.



## Discussion and conclusions

None of the two calving seasons appeared to be technically or economically better than the other. Only climatic conditions (good grass growth in summer) and the farmer's holiday choices will help him choose among the two options. Compact calvings lead to a strong monthly work specialization. But they require the production of heifers calving at 24 months. With Holstein high genetic merit cows a system based on a 3-month period appeared more repeatable than a 2-month one. This experiment confirmed that OAD milking can be achieved over the whole lactation on grass based systems with low feeding costs even for high genetic cows. No consequence on animal welfare was recorded (Brulé *et al.*, 2003; Tucker, 2007). A good initial cell-count situation is compulsory. The decrease in the working time reached 17% but more important for the farmer is the increased flexibility of OAD milking. Turning to OAD milking, at least in early lactation, can make it easier to reach the grouping targets.

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# Maintaining grassland-based production in regions with low rainfall and dry soils: Controlled Maine-Anjou Origin Appellation (AOC Maine-Anjou)

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## Abstract

In regions with low rainfall, dry soils and poor water retention, cattle farmers are forced into maize silage production on irrigated areas in order to maintain forage autonomy, at the expense of grasslands. In addition, with the liberalisation of the European agricultural market, cattle farming activities are disappearing, replaced by cash cereal crops. In this context, one way for farmers to continue grassland-based cattle farming is to market their products with a label of quality which guarantees a certain proportion of grass in the diet. However, grassland management and its consequences on forage autonomy are poorly understood. Ninety-three farms in the Loire Region (France) with beef cattle fed mainly grass (Controlled Maine-Anjou Appellation – AOC Maine-Anjou) were selected. Grassland management was studied using a multivariate approach. The results showed a high proportion of grass in the forage produced (89% grass as opposed to 11% maize silage). The management of these grasslands is closely monitored: (1) 60% are temporary grasslands, (2) seed species and mixtures are chosen to maximize grass yield, (3) the grazing period is maximized (9 months on average). This makes grass forage autonomy possible for 74% of farms investigated based on two strategies: a nearly entire grass-based system or a grass-maize-straw based system.

Keywords: grassland management, dry area, beef cattle, specifications, forage autonomy

## Introduction

In European regions with low rainfall, dry soils and poor water retention, beef-cattle farming is often, paradoxically, one of the main types of agricultural production (Sarzeaud, 2006). Beef-cattle farmers have developed cropping plans composed of grasslands, maize silage and cereals to maximize feed autonomy. Since 2003 and the liberalisation of the European agricultural market, many other agricultural productions, such as cash crops, have been developed at the expense of cattle production. This has led to a large decrease in the proportion of grasslands. However, many studies have shown that maintaining grasslands should be an objective, mainly for environmental reasons (Le Gall *et al.*, 1997). One way for farmers to continue grassland-based cattle farming is to market their products with a label of quality which guarantees a certain proportion of grass in the diet. But in such cases, cattle farmers need to maintain or/and improve the autonomy of their forage system based mainly on grass, in a context of climate change (more frequent droughts). Many adjustments of grassland management are possible by varying: proportion of grassland in forage crops, livestock units per ha of grass, storage methods of grass, grass species and varieties sown, length of grazing period, (Lemaire and Pfimlin, 2007). Nevertheless, these adaptations and their consequences in terms of forage autonomy are poorly known. The objective of our study was to describe grassland management and its consequences on forage autonomy using a case study: Controlled Maine-Anjou Appellation (AOC Maine-Anjou) which is a beef cattle production based on grass (at least 1 ha of grassland per beef cow) in a region with low rainfall and dry soils (Loire region, west of France).



## Materials and methods

We investigated beef cattle management in AOC Maine Anjou farms. The AOC farms were selected for the size of the herd (at least 20 beef cows) and for the number of animals sold in the AOC market (at least 2 culled cows and/or steers in 2005 or 2006). In this way, 158 farms were chosen and 92 from this sample were studied in November 2006. The questionnaire was mainly composed of closed questions. The following themes were investigated: description of the farming system, cropping plan in 2006, grassland management and forage management. Firstly, data were analysed using descriptive analysis to qualify grassland management in the whole data set. Then, 4 groups of farms were constituted on the basis of their forage autonomy and the level of grass in the forage produced on the farm: G1 = self-sufficient / > 80% of grass, G2 = self-sufficient / < 80% of grass, G3 = dependent / > 80% of grass, G4 = dependent / < 80% of grass. The 4 groups were compared using t-tests to analyse grassland management strategies.

## Results and discussion

The farms were composed of  $52 \pm 22$  beef cows ( $99 \pm 39$  LU). 45% of the farms were specialized in beef cattle production. Dairy cattle, poultry, pig and sheep/goat were observed in 27%, 20%, 6% and 4% of the farms, respectively. The total agricultural area (TAA) and forage crop area were  $100.9 \pm 47.6$  ha and  $80.5 \pm 38.3$  ha, respectively.

The proportion of grasslands was high ( $72.1 \pm 35.3$  ha,  $73.8 \pm 16.1\%$  of the TAA), compared to average figures for beef cattle farms of the region (60% of the TAA; Sarzeaud, 2006). These results confirm the impact of the AOC specifications on the cropping plan: the grassland area per beef cow was  $1.44 \pm 0.57$  ha. Temporary grasslands (TG) represented  $60.9 \pm 31.3\%$  of grassland. The TG species were chosen to increase the productivity and the longevity of grasslands and to spread grass production throughout the season. The TG were composed of  $29 \pm 34.3\%$  monoculture (Italian ryegrass (IR), perennial ryegrass (PR), fescue (F), cocksfoot (C), hybrid ryegrass (HR)),  $29 \pm 36.9\%$  2 species-associations (PR-white clover (WC), F-C, F-WC, and HR-WC),  $9 \pm 23.9\%$  3 species-associations (mainly PR-F-WC),  $18 \pm 35.1\%$  multispecies TG and  $15 \pm 29.4\%$  old TG. The grazing season lasted  $251 \pm 31.4$  days. 81% of farmers gave hay or haylage to the animals from June to the end of the grazing season in addition to the grazed grass.

100%, 46% and 44% of farmers stored grass as hay, haylage and silage, respectively. The amount of grass forage produced on farms was  $1.69 \pm 0.85$  DM t / LU. Maize silage, straw and cereal silage were produced in 62, 43 and 12% of the farms, respectively. The amount of forage produced was  $2.41 \pm 1.00$  DM t / LU. 44% of farms were self-sufficient for forage, 20% were self-sufficient for grass and bought mainly straw. The 26% remaining were dependent on grass and all bought hay while 4 bought maize silage and 10 straw. Finally, the amount of stored forage (produced and purchased) was  $2.67 \pm 1.04$  DM t / LU. This amount corresponds to regional figures (Sarzeaud, 2006) and can be explained by the length of the grazing season and the use of fibrous concentrate in a high proportion in the diet.

Farmers have developed two strategies for producing forage: a nearly entirely grass-based system (G1 and G3) or a grass-maize-straw-based system (G2 and G4) (Table 1). The G1 and G3 farms had smaller TAA, higher proportions of forage crops in the TAA and grasslands in the forage crops area, than the G2 and G4 farms (Table 1). These differences can be explained by the production of young bulls and milk in G2 and G4 farms. Types of grasslands, length of grazing season, livestock density on grasslands did not differ between strategies. The G1 and G3 farmers tended to mow once more (haylage) than the G2 and G4 farmers (Table 1). This additional mowing could partly induce the greater amount of grass forage produced in the G1 and G3 farms. This extra-mowing could be explained by a higher grass yield in the grasslands

(perhaps due to greater fertilization, data could not be analysed in our study) and/or to favourable local climatic and soil conditions.

Table 1. Characteristics of farms according to their level of forage autonomy (self-sufficient or dependent) and the strategy developed to produce the forage (grass or grass-maize-straw).

	Self-sufficient		Dependent	
	Grass G1 (n = 20)	Grass-Maize G2 (n = 30)	Grass G3 (n = 19)	Grass-Maize G4 (n = 23)
n = 92				
Total surface area, ha	99.9ab	104.8a	80.6b	110.9a
Forage crops, % of total agricultural area	83.3a	77.2b	84.6a	79.1ab
Grasslands, % of forage crops	93.0a	86.2b	99.2c	84.1b
Permanent grasslands, % grassland area	37.9	38.8	32.9	43.5
Livestock density on grasslands, LU/ha of grasslands	1.35	1.52	1.56	1.51
Grassland monocultures, % of temporary grasslands	31.9a	23.9abc	11.8b	28.4ac
Multispecies grasslands, % of temporary grasslands	37.9a	51.4ab	54.4ab	63.6b
Old grasslands, % of temporary grasslands	15.2abc	8.1ac	28.5b	3.6c
Only grazing area, % of grassland area	47.3	46.9	44.3	52.2
Total area of mowed grassland, ha	51	41	51	41
Forage autonomy, %	100a	100a	75b	84c
Forage produce, DM t/LU	2.43ab	2.66a	1.97b	2.44ab
Including conserved grass produce, DM t/LU (%)	2.27a (94)	1.60b (62)	1.84b (94)	1.25c (51)
Total forage, DM t/LU	2.43	2.66	2.66	2.91
Including total conserved grass, DM t/LU (%)	2.27a (94)	1.60b (62)	2.10a (71)	1.43b (43)

<sup>a, b, c</sup>: significant difference  $P < 0.1$ .

In the nearly entirely grass-based system, the self-sufficient farms (G1) tended to be greater ( $P = 0.20$ ) and to have a lower livestock density on the grasslands ( $P = 0.12$ ), than the dependent farms (G3). The level of forage crops in the TAA was similar between G1 and G3 farms. The proportion of monocultures (mainly IR and C) in TG was higher ( $P = 0.05$ ) and the proportions of multispecies and old TG tended to be lower ( $P = 0.20$  and  $P = 0.12$ , respectively) in the G1 farms (Table 1). These results could mean that G1-farmers aim to maximize the yield of their grasslands at the expense of longevity. A higher level of fertilization might confirm this interpretation. The self-sufficient (G2) and dependent (G4) grass-maize-silage-based systems had similar cropping plans and similar grassland management. Our results only partially explain the difference of grass forage produced between the 2 groups. The presence of dairy cattle, the level of fertilization and the local climatic and soil conditions may explain the difference observed and will be investigated in a future study.

## Conclusions

In 2006, a drought year, being self-sufficient for forages was possible in grass-based systems situated in regions with low rainfall and dry soils. Many adaptations have been shown and others need to be studied (e.g. fertilization). Farms often purchase fibrous concentrate. It would be interesting to enlarge this study to include concentrate and to analyse total feed autonomy.

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# Ruminal fermentation pattern in dairy cows as affected by herbage proportion in the diet and feeding level

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## Abstract

Six stall-fed Holstein dairy cows were used in a 4×4 Latin square design to study the effect of the proportion of fresh perennial ryegrass and maize silage (mixed with 13% soyabean meal, DM basis) in the diet and the effect of feeding level on ruminal fermentation pattern. The ratio herbage:maize silage in the diet was 25:75, 50:50, 75:25 and 50:50 for treatments 1, 2, 3 and 4, respectively. Feeding level was 100% *ad libitum* for treatments 1, 2 and 3, and 70% *ad libitum* for treatment 4. Cows were fed herbage in the morning and maize silage in the evening. Mean ruminal pH and VFA were only slightly affected by treatments. Molar proportions of VFAs were affected by herbage proportion in the diet only in the morning. Post-prandial variation of the molar proportions of VFAs differed between morning (herbage) and evening (maize silage) meals.

Keywords: ruminal fermentation, dairy cow, forage type, diet composition, feeding level

## Introduction

In dairy systems, during periods of transition or herbage shortage, cows generally graze during the day and can be housed and supplemented with maize silage at night. The effect of such within-day variation of forage type and/or quantity on ruminal fermentation has not been investigated. This study aims to determine the influence of different herbage:maize silage ratios in the diet and of feeding level on the stability of ruminal fermentation in dairy cows fed herbage during the day and maize silage at night as in grazing situations.

## Materials and methods

Four treatments were compared. In the first three treatments, the diet consisted of (DM basis) 25 (H<sub>25</sub>), 50 (H<sub>50</sub>) or 75 (H<sub>75</sub>)% of freshly cut perennial ryegrass, cows being fed at 100% *ad libitum*. In the fourth treatment (H<sub>50/70</sub>), cows were fed the H<sub>50</sub> diet but only at 70% *ad libitum*. The other component of the diet (the supplement) was a mixture comprising 87% of maize silage and 13% of soyabean meal on a DM basis. The experiment was carried out on 6 multiparous Holstein dairy cows in mid lactation fitted with a rumen canula, according to a 4×4 Latin square design. Each period lasted 14 days.

Herbage was cut once daily early in the morning and given in two meals at 08:30 and 11:30 h. The supplement was given once daily at 17:00 h. Refusals of herbage and supplement were removed and weighed at 16:30 and 08:00 h, respectively. Cows were milked at 06:30 and 17:00 h.

Intake and milk production were measured each day and milk fat content was determined from day 10 to day 14. Ruminal fermentation pattern was studied on day 13 by sampling ruminal liquid in the ventral sac at 07:00, 08:00, 11:00, 14:00, 16:00 and 20:00 h. Mean daily values were calculated by weighing the observed values by the duration between samplings.

Average data per cow and period were subjected to variance analysis taking into account the effects of cow, period and treatment. Orthogonal contrasts were designed to test the effect of feeding level (FL, H<sub>50</sub> vs H<sub>50/70</sub>), and the linear (HP<sub>LIN</sub>) and quadratic (HP<sub>QUA</sub>) effects of the

herbage proportion in the diet. The effect of time and their interactions were also tested considering repeated measurements.

## Results and discussion

Total DM intake was lower than expected in H<sub>75</sub> because of herbage refusals (Table 1). Actual herbage proportions in the diet were 0.24, 0.47, 0.69 and 0.47 for treatments H<sub>25</sub>, H<sub>50</sub>, H<sub>75</sub> and H<sub>50/70</sub>, respectively. The CP content and *in vitro* OM digestibility values were 175 g kg<sup>-1</sup> DM and 0.79 for the herbage, and 131 g kg<sup>-1</sup> DM and 0.75 for the supplement, respectively. Milk production was not affected by herbage proportion in the diet and decreased by 3.8 kg when decreasing feeding level ( $P < 0.01$ ). Milk fat content was not affected by treatments (Table 1).

Excepting ammonia, mean ruminal fermentation parameters were only slightly affected by treatments (Table 1). Ammonia concentration linearly increased with increasing herbage proportion in the diet and increased with decreasing feeding level ( $P < 0.01$ ), as previously observed by Stockdale (1994) on similar type of diet. Ruminal pH tended to increase with decreasing feeding level. Iso-acids (IsoC<sub>4</sub> + IsoC<sub>5</sub>) proportion decreased and VFAs concentration tended to increase with increasing herbage proportion in the diet, despite lower total DM intake. This result may be partly explained by the low number of sampling times during the night after feeding the supplement. Higher ruminal VFA concentration in H<sub>25</sub> should be expected with more regular sampling times throughout the 24-h period. There was no treatment effect on the molar proportions of acetate, propionate and minor acids (C<sub>4</sub> + C<sub>5</sub>) nor on the ratio (C<sub>2</sub> + C<sub>4</sub>)/C<sub>3</sub>, which supports the absence of variation of milk fat content.

Table 1. Effects of herbage proportion in the diet and of feeding level on intake, milk production and mean ruminal fermentation parameters in lactation dairy cows.

Parameter	Treatments				s.d.	Effects ( $P < $ )		
	H <sub>25</sub>	H <sub>50</sub>	H <sub>75</sub>	H <sub>50/70</sub>		HP <sub>LIN</sub>	HP <sub>QUA</sub>	FL
Herbage DM intake (kg d <sup>-1</sup> )	4.5	8.7	11.3	6.3	0.53	0.001	0.02	0.001
Total DM intake (kg d <sup>-1</sup> )	18.2	17.9	16.1	12.9	0.46	0.001	0.01	0.001
Milk production (kg d <sup>-1</sup> )	29.8	27.9	28.1	24.1	1.71	ns	ns	0.01
Milk fat content (g kg <sup>-1</sup> )	37.5	38.8	38.1	39.3	1.20	ns	ns	ns
Ammonia (mg N-NH <sub>3</sub> L <sup>-1</sup> )	67.2	77.0	94.5	104.1	14.49	0.01	ns	0.01
pH	6.20	6.16	6.11	6.35	0.180	ns	ns	0.10
VFA (mmol L <sup>-1</sup> )	95.8	100.2	105.3	91.2	8.76	0.10	ns	ns
Acetate (%)	63.2	64.1	64.3	64.5	1.42	ns	ns	ns
Propionate (%)	20.0	19.6	20.5	19.6	1.34	ns	ns	ns
Butyrate (%)	12.1	12.3	11.3	12.0	0.62	0.08	0.09	ns
Iso-acids (%)	2.74	2.34	2.18	2.47	0.267	0.01	ns	ns
Minors (%)	1.99	1.72	1.62	1.40	0.460	ns	ns	ns
(C <sub>2</sub> +C <sub>4</sub> )/C <sub>3</sub>	3.84	4.03	3.79	4.01	0.354	ns	ns	ns

Time effect was significant for all ruminal parameters ( $P < 0.001$ ) as illustrated in Figure 1. The interaction time  $\times$  treatment was significant only for ammonia concentration, acetate, butyrate and minor acids proportions ( $P < 0.001$ ), and for VFAs concentration ( $P < 0.03$ ).

The evolution of ruminal fluid composition between 0 and 3 h post-feeding the herbage (morning) or the supplement (evening) shows that feeding both types of feed decreased ruminal pH, increased VFAs and ammonia concentration (Figure 1). However, it is noteworthy that post-prandial variations of the molar proportions of VFAs strongly differed between herbage and supplement meals, whatever the treatments and amount offered to cows. Feeding herbage reduced acetate and iso-acids proportions while strongly increasing propionate proportion. Feeding supplement increased butyrate, iso-acids and minor acids proportion while reducing propionate proportion.

Molar proportion of the VFAs were mainly affected by the herbage proportion in the diet early in the morning before feeding, which may be linked to higher energy availability for ruminal microbes during the night for cows receiving high level of supplement in the evening. Similar results were found by Graf *et al.* (2005) when supplementing grazing dairy cows with 6 kg DM of maize silage in the evening.

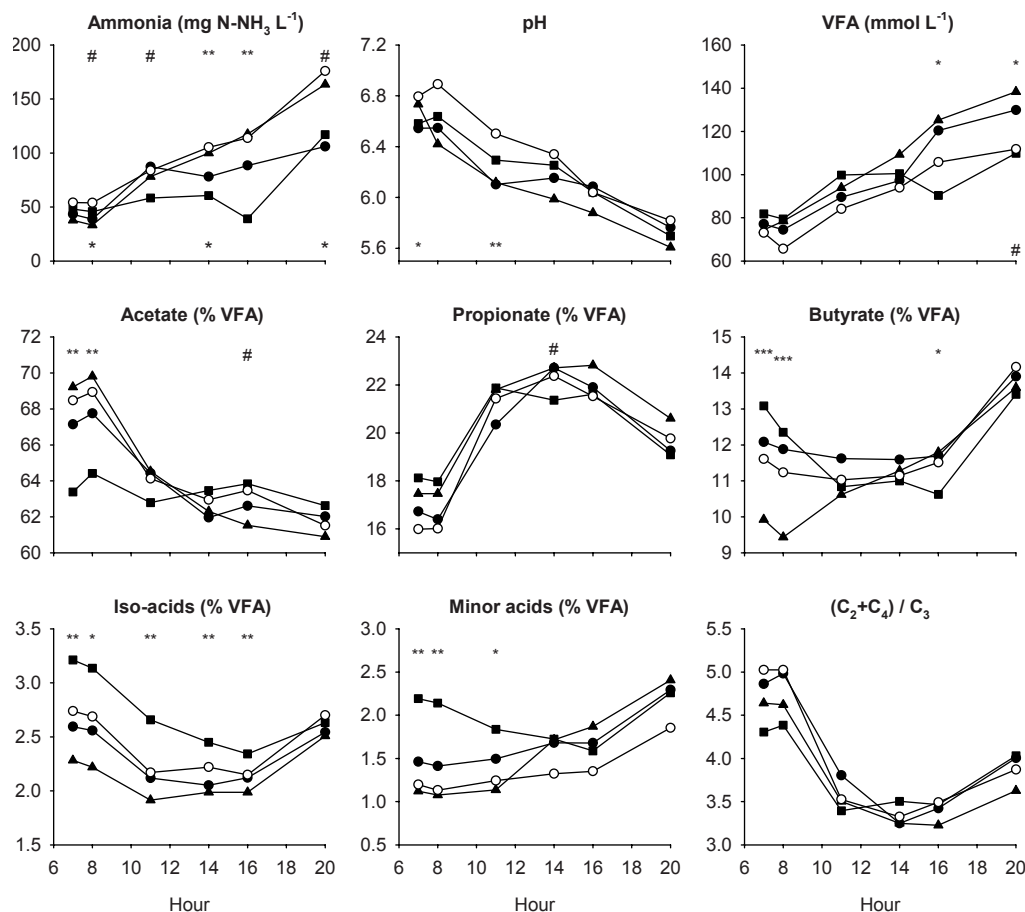


Figure 1. Effects of herbage proportion in the diet (■ 25%, ● and ○ 50%, ▲ 75%) and of feeding level (full symbols: 100% *ad libitum*, empty symbols: 70% *ad libitum*) on the ruminal fermentation pattern in lactating dairy cows. Herbage proportion effect: top of each graph. Feeding level effect: bottom of each graph (#  $P < 0.10$ , \*  $P < 0.05$ , \*\*  $P < 0.01$ , \*\*\*  $P < 0.001$ ).

## Conclusions

When fresh herbage is fed in the morning and maize silage-soyabean meal in the evening, large variations in herbage proportion in the diet and in feeding level only slightly affected the milk fat content and mean daily ruminal fermentation parameters. Despite large differences in the post-prandial variations of the molar proportions of VFAs according to the forage fed, herbage proportion in the diet affected molar proportion of VFAs only early in the morning. It can be concluded that the conditions of ruminal digestion are fairly stable in a wide range of forage-based feeding strategies.

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# Short time eating rate by goats and cows of long and chopped grass silage, harvested at three stages of growth

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## Abstract

Short-term eating rate of different silages was studied as a measure of forage palatability. The same grass silages were fed to goats in late gestation, goats in early lactation and dairy cows in early lactation, aiming to evaluate the preferences by the different groups of animals. The silages were harvested at an early, normal and late stage of maturity, and fed long or chopped (225 mm and 19 mm median particle length, respectively). Each trial included three 6 x 6 Latin squares. One square was done each day with three 15-minute periods in the morning and three in the afternoon. The feed rations for each period consisted of 0.4 kg and 4 kg for goats and cows, respectively. Chewing activity and eating time were measured using IGER Behaviour Recorders. Higher feed residues were recorded from the late harvested silage, long and chopped, and from medium harvested long silage, than from the other qualities. Short-term eating rate (DM intake min<sup>-1</sup>) decreased with increasing growth stage of grass silage, and was significantly greater for chopped than long silage at all harvesting times for all groups of animals.

Keywords: silage, growth stage, chopped, eating rate, goats, cows

## Introduction

According to Van Soest (1994) the eating rate of a diet (kg DM min<sup>-1</sup>) depends on the fibre content, palatability, the animal's hunger and the metabolic state of the animal. Forages with small particle size are consumed at a high eating rate, while coarse forage with high fibre content is eaten more slowly. The eating rate of forages can therefore be increased by chopping the forage (Nørgaard, 2003). For grass silage rations, other palatability factors like fermentation quality may also influence eating rate (Garmo *et al.*, 2007). One of the objectives of this experiment was to investigate the effect of stage of maturity and chop length of silage on the eating rate of early lactating cows and goats, and goats in late gestation. Goats in lactation have a higher feed requirement than pregnant goats (Morand-Fehr and Sauvant, 1989), so another objective was to investigate whether this change of physiological stage influences eating rate. Garmo *et al.* (2007) found significant correlations between short time eating rate and 24-hour eating rate for lactating cows, and between short time eating rate and daily feed intake during a two-week period. Because short-time eating rate is a rapid, simple, and therefore cheap test of feed palatability, we chose that method for this study, and assumed that different forage qualities would be ranked in the same order as in a long-term intake study.

## Materials and methods

Short-term eating rates were tested on: 1) goats in late gestation, 2) cows in early lactation, and 3) goats in early lactation. The grass silage, dominated by timothy, was harvested in May and June 2006, and baled using an Orkel GP 1260 roundbaler with 20 fixed knives. At early, normal and late stage of maturity the energy content of fresh herbage analysed by NIRS was 1.00, 0.96 and 0.87 FEm kg<sup>-1</sup> DM<sup>-1</sup>, and mean stage by weight 2.35, 2.69 and 3.12, respec-

tively. The silages from all three harvesting times were chopped to two different lengths: (1) no further chopping, (2) finely chopped (Serigstad RBK 1202 + epple Blasius 940). Each trial included three 6 x 6 Latin squares. One square was done each day with three test periods in the morning and three in the afternoon. Each test period lasted for 15 minutes, with a 10-min interval between each period. The feed rations for each period consisted of 0.4 kg and 4 kg for goats and cows, respectively. Residues were weighed and analysed. Chewing activity and effective eating time were measured using IGER Behaviour Recorder (Rutter *et al.*, 1997). Eating time was also measured manually as total time eating. The test animals had free access to silage during the night and between morning and afternoon test periods, except for two hours before the tests started. Concentrates were given at least two hours before the test start. Daily concentrate allowances were 0.4 kg, 0.9 kg and 9 kg, and mean body weights were 63.3 kg, 60.4 kg and 647.5 kg for the pregnant goats, lactating goats and lactating cows, respectively. The daily milk yields of lactating goats and lactating cows were 4.3 and 40 kg, respectively.

Table 1. Chemical content of the silage (g kg<sup>-1</sup> DM<sup>-1</sup> if nothing else given).

	Early harvested silage	Normal harvested silage	Late harvested silage
DM g kg <sup>-1</sup>	303	277	314
Crude protein	166	135	115
NH <sub>3</sub> -N g kg <sup>-1</sup> TN <sup>-1</sup>	90	98	115
pH	4.3	4.1	4.4
NDF	452	533	613
ADF	288	337	371
Ash	113	66	67
ADL	42	44	60
Crude fat	42	37	29
Acetic acid <sup>1</sup>	8.7	8.0	7.9
Lactic acid	46.9	45.3	45.0
Formic acid	13.2	13.7	9.4
Ethanol	7.4	27.0	10.7

<sup>1</sup>Propionic acid and butyric acid were not detected.

## Results and discussion

The fermentation quality of the silages was good, and similar for the three harvesting times (Table 1). Median particle length of the offered silages, measured by hand sorting, were for long and finely chopped silage 163 and 19 mm, 192 and 21 mm, and 325 and 18 mm, for early, normal and late harvested silage, respectively. Intake and eating rates are presented in Table 2. Higher feed residues were recorded from the late harvested silage, long and chopped, and from medium harvested long silage, than from the other qualities. Early harvested and medium harvested chopped silage were almost completely ingested during the test period in all trials. The NDF and protein content in the feed residues indicated that feed selection during the 15-minute periods was negligible for all animal groups. Effective eating time (min) was significantly shorter for finely chopped silage compared to long silage. Short time eating rate (g DM min<sup>-1</sup> and g NDF min<sup>-1</sup>) decreased with increasing growth stage of silage for all groups of animals. For example, for lactating goats the mean effective eating rate for early, normal and late harvest time was 24, 12 and 6 g DM min<sup>-1</sup>. Chopping the silage increased eating rate at all harvesting times, except for the manually measured eating rate at the late harvesting time. The former is in agreement with Garmo *et al.* (2007) who also found that the relative effect of the best quality forage was higher in a short term eating rate test compared with 24-hour eating rate. The physiological stage of the goats did not affect the general eating rate ( $P = 0.8$ ) or dietary preference, as also stated by Morand-Fehr (2003). This may be caused by the goats' hunger during the tests or the fact that some individual goats had a poor feed



intake during the tests. Due to higher energy demand by lactating goats than by dry goats a higher eating rate was expected for lactating goats. Any such difference could, however, not be detected in this short-time eating rate study.

Table 2. Feed intake, eating time (min), eating rate (g DM min<sup>-1</sup>, g NDF min<sup>-1</sup>) by pregnant and lactating goats and lactating cows (LS means). ( $P < 0.05$ ).

Harvesting time	Early		Normal		Late		SEM	Effect ( <i>P</i> )		
Chopping length	Long	Fine	Long	Fine	Long	Fine		Time <sup>1</sup>	Chop <sup>2</sup>	TxC <sup>3</sup>
<b>Pregnant goats:</b>										
Feed intake, g DM	111	111	73	109	58	49	6.1	< 0.001	ns	0.001
Eating time effective	6.0	4.6	8.7	5.7	9.7	4.5	0.7	0.007	< 0.001	0.02
Eating time manually	9.6	7.7	14.7	8.3	15.0	14.9	0.6	< 0.001	< 0.001	< 0.001
Eating rate effective, DM	19	27	8	20	6	9	1.1	< 0.001	< 0.001	0.02
Eating rate manually, DM	14	19	5	14	4	3	1.1	< 0.001	< 0.001	< 0.001
Eating rate effective, NDF	9	12	5	11	4	5	0.6	< 0.001	< 0.001	ns
<b>Lactating goats:</b>										
Feed intake, g DM	110	104	50	91	26	21	6.3	< 0.001	0.05	0.002
Eating time effective	6.5	3.8	7.2	5.1	5.5	2.2	0.6	0.001	< 0.001	ns
Eating time manually	10.4	7.1	15.0	10.1	15.0	15.0	0.6	< 0.001	< 0.001	0.003
Eating rate effective, DM	19	30	8	16	5	8	1.3	< 0.001	< 0.001	0.01
Eating rate manually, DM	13	19	3	11	2	1	1.1	< 0.001	< 0.001	< 0.001
Eating rate effective, NDF	8	14	5	9	3	5	0.7	0.05	< 0.001	ns
<b>Lactating cows:</b>										
Feed intake, g DM	1192	1203	842	1081	234	99	32.8	< 0.001	ns	< 0.001
Eating time effective	11.9	8.6	13.7	11.2	7.9	2.4	0.6	< 0.001	< 0.001	0.03
Eating time manually	12.3	9.0	14.8	12.1	15.0	15.0	0.3	< 0.001	< 0.001	< 0.001
Eating rate effective, DM	103	142	62	98	24	19	3.4	< 0.001	< 0.001	< 0.001
Eating rate manually, DM	100	136	58	92	16	7	3.6	< 0.001	< 0.001	< 0.001
Eating rate effective, NDF	46	64	33	53	15	11	1.9	0.03	< 0.001	0.06

<sup>1</sup>Harvesting time <sup>2</sup>Chopping length <sup>3</sup>Harvesting time x chop length interaction.

## Conclusion

Chopping of silage, and harvesting at an early state of maturity, increased short time eating rate for all groups of animals. The feed ranking by cows and goats seemed to be similar. Neither differences between animal species in feed selection capability, nor differences due to physiological stage of the animal could be detected in this short-time eating rate study.

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# Farmers' perspectives for forage-based dairy production. A Review

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## Abstract

Consumers are increasingly aware of health aspects of food products. Milk fatty acid (FA) composition is largely influenced by animal feed. Feeding systems have greatly changed during the last decades, as have milk production levels of dairy cows.

A survey of milk of Dutch farms revealed large regional differences in the FA profile and conjugated linoleic acid (CLA) concentration of milk, which could be related to differences in diet and feeding system as main explanatory variable. Regions with a high amount of forage maize cultivation produced milk with less unsaturated FA than regions with predominantly grass. Milk from farms where cows had unrestricted access to pastures generally had higher CLA concentrations than when cows had restricted access, and much higher than when cows were kept indoors on a silage diet. In 2007 the two major Dutch dairy cooperatives launched grass-based dairy products. Farmers receive a premium price, but future success will depend on farm economics and on continued consumer willingness to pay a higher price for these products.

Keywords: milk price, grass-based dairy, marketing, dairy image, healthiness

## Introduction

Consumers are increasingly aware of health aspects of food products. Milk fatty acid (FA) composition is largely influenced by animal feed. Feeding systems have greatly changed during the last decades, as have milk production levels of dairy cows. There is a trend in many countries with intensive dairy systems, including The Netherlands, of keeping cows more indoors. However, it is undesirable that concentrations of beneficial FA in dairy products are nowadays lower than before because of changes at farm level. Because FA composition measurements are not routinely done, this has long remained largely unnoticed by the industry and by the general public, but recent research outcomes have generated much interest and induced changes.

Surveys of milk of hundreds of Dutch farms were carried out at various times during the season with the aim to monitor milk FA pattern, in particular the CLA (conjugated linoleic acid) concentration, and relate it to region, soil type, feeding system, herd characteristics and milk composition to obtain more insight in causes of regional and temporal patterns of milk FA composition.

## Materials and methods

Farms were surveyed in various regions in The Netherlands. In one study, milk was collected monthly of all farms for each of four regions (A-D) by pooling commercial tank milk samples during 12 months in 2000/2001. In another study, milk samples were collected from farms in various regions and information about farm size, soil type, herd characteristics and feeding regime was obtained from questionnaires. Milk fat, protein and lactose data were obtained from the milk testing station and samples for FA analyses were stored at -18° C until analyses for CLA concentration in the milk fat (Elgersma and Wever, 2005). Regression analysis was used to relate the various parameters as explanatory variables to model CLA concentration and production.



## Results and discussion

In the first study, clear temporal and regional differences in milk FA composition occurred. The CLA concentrations are shown in Figure 1. As there is a linear relation between concentrations of CLA and total unsaturated FA, CLA can be used as an indicator of the FA profile (Elgersma *et al.*, 2006). CLA concentrations were lower during the indoor season (Nov-Apr) than during the growing season (May-Oct). The CLA concentration from Nov-April was on average 0.3 g/100 g milk fat, while the highest levels were recorded in August (0.8 g/100 g). However, there were very pronounced regional differences. The two regions A and B, with mainly sandy soils and a high amount of forage maize cultivation, produced milk with lower CLA and unsaturated FA concentrations than regions C and D, with predominantly grass. Differences were most pronounced during the growing season.

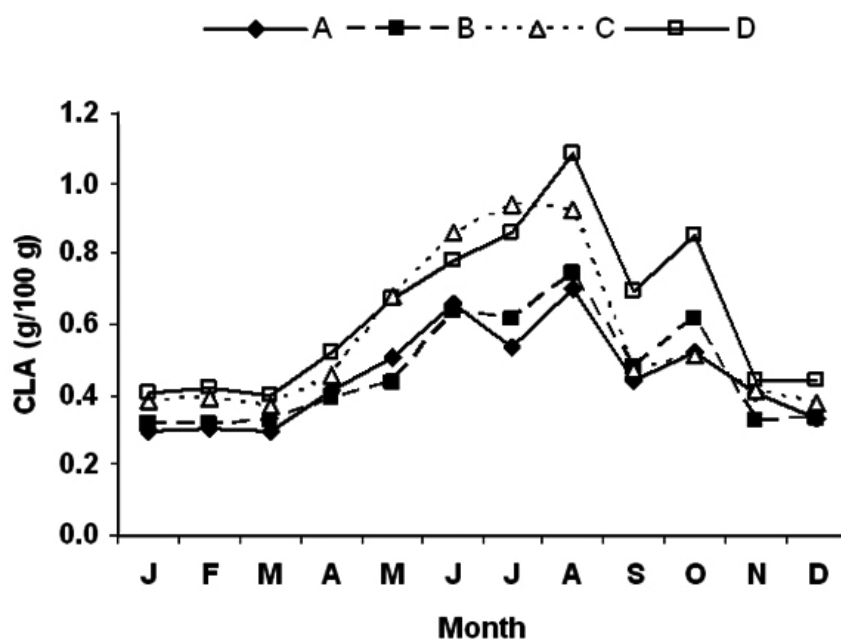


Figure 1. Concentrations of conjugated linoleic acid (CLA) in milk fat from milk of Dutch dairy farms in 4 regions (A-D) produced in the period August 2000-July 2001.

In the second study, milk CLA concentrations in the region of Noordelijke Friese Wouden were measured and related to feeding regime and grazing management. Milk from farms where cows had unrestricted access to pastures during summer generally had higher CLA concentrations than when cows had restricted access, and much higher than when cows were kept indoors on a silage diet (Figure 2). In February there was no such relation. Regression models showed that the variation in CLA concentration in February was explained most by milk fat content, but in September by urea content. No relation with milk protein concentration was found. The average CLA concentrations in this region were higher than reported earlier for Dutch milk (Figure 1). Within the group of farms with unrestricted stocking there was a wide range in CLA concentrations. In summary, these studies revealed large temporal and regional differences in the CLA concentration of milk that could be related to differences in diet and feeding system as main explanatory variable.

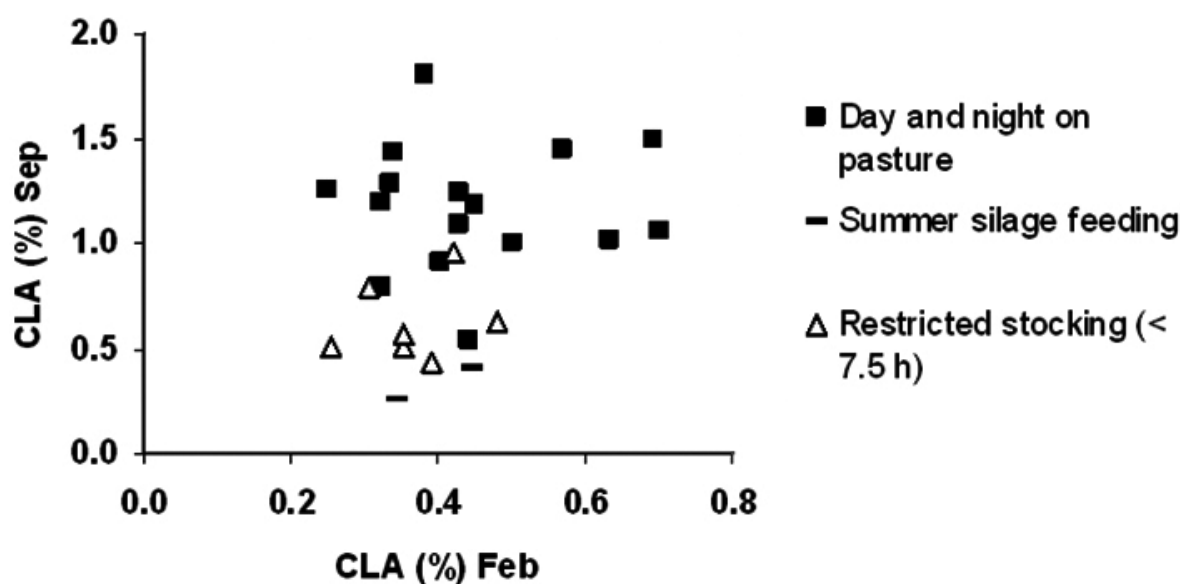


Figure 2. CLA concentrations in milk sampled in February and September 2006 of farms with different feeding regimes.

From spring 2007 onward, the two major Dutch dairy cooperatives (Campina and Friesland Foods) have followed earlier initiatives of smaller dairy producers, and launched grass-based dairy products. All their liquid milk sold in The Netherlands is now labelled as ‘weidemelk’ (i.e., pasture-based milk). Campina also imposes changes on the concentrate feed composition fed to cows of the farmers delivering milk to the plants that produce these products. Farmers now receive a premium price, but future success will depend on farm economics and on consumer willingness to pay a higher price for these products.

## Conclusions

- There are large regional differences in the conjugated linoleic acid (CLA) concentration of milk
- These can be related to diet and feeding system as main explanatory variable
- There is thus scope for farmers to produce milk with a modified fatty acid composition
- Dutch farmers now receive a premium price
- Dutch dairy cooperatives have launched grass-based dairy products
- Future success will depend on farm economics and consumer willingness to pay a higher price for these products

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## Triticale and mixtures silages for feeding dairy cows

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### Abstract

In areas where the conservation of water resources is becoming of strategic importance, farmers and advisers are requiring silages made without irrigation and pesticides. We evaluated in this study the agronomic and zootechnical performances of winter crops harvested as silage in spring. To determine the effects of the presence of legumes (simple or complex mixture) compared to pure cereals, three crops were compared: a pure triticale (*Triticosecale* W.) crop (T1), a triticale and forage pea (*Lathyrus sativus* L.) mixture (T2), and a complex mixture of triticale, pea, oat (*Avena sativa* X.) and vetch (*Vicia sativa* L.) (T3). The crop trials were conducted in large fields (2 ha each), on a low-input management basis (no pesticides, mineral nitrogen provided only to T1 treatment). The three silages were offered *ad libitum* to 24 mid-lactation dairy cows in a replicated 3x 3 Latin square design. Periods were 3 weeks long. Intake and animal performances were measured. The forages yielded 8.9, 8.1 and 7.4 t DM ha<sup>-1</sup> respectively for T1, T2 and T3 treatments. The effect of treatment on forage intake and milk production were significant. Daily intake were 9.0 kg, 10.1, 10.9 and milk production were 19.6 kg, 20.6, 21.4 respectively for T1, T2 and T3. Cereal and mixtures silages, harvested before the summer drought should provide useful forage without needing irrigation.

Keywords: triticale, pea, mixture, dairy cows, silage, water

### Introduction

Under north-west European conditions, whole-plant maize silage is the main ingredient in the diets of dairy cows, especially during the winter, because of its yield potential and feeding value. Nevertheless, maize production could be limited in some areas because summer rainfall is insufficient, so it has to be supplemented by irrigation which then generates an important agricultural water consumption. Winter cereals harvested as silage could be of great interest because they allow farmers to escape the summer drought. If devoted to be fed to ruminants they can be produced with a low level of inputs, which is also favourable in terms of sustainable agriculture. In this study we evaluated the agronomic and zootechnical performances of three winter pure-cereal or mixture crops harvested as silage in spring. Triticale was chosen as the basic component of this experiment because of its hardiness and high-yield potential under limiting conditions and its quality parameters (Emile *et al.*, 2007). The objective of this study was to evaluate the interest of adding legumes to triticale from both a crop yield and an animal performance point of view.

### Materials and methods

The crops were a pure triticale (*Triticosecale* W.) crop (T1), a binary mixture with triticale and forage pea (*Lathyrus sativus* L.) (T2), and a complex mixture of triticale, pea, oat (*Avena sativa* X.) and vetch (*Vicia sativa* L.) (T3). The crops were sown on 7-9 November 2005 in

Lusignan (0°08'35''E, 46°40'39''N, Poitou-Charentes, France) on 2-ha plots each. The sowing densities (grains m<sup>-2</sup>) were 220 for triticale (except 110 for T3), 110 for oat, 17 for pea and 20 for vetch. No pesticides were used on the crops. The two mixtures (T2 and T3) including legumes were not fertilized, whereas 80 kg N were provided for the pure triticale crop. The crops were harvested as silage on 6-8 June 2006 at the triticale late milk-early dough grain stage. Farm direct costs, including pesticides, fertilizers and seeds were recorded. Direct harvest was performed using a rotary head harvester. Storage and distribution conditions of both silages were strictly comparable. Dry matter (DM) yield was evaluated at the date of harvest on four 1 m<sup>2</sup> plots of each crop. The three silages were offered *ad libitum* to 3 groups of 8 mid-lactation dairy cows (50% primiparous cows, 27.0 kg milk day<sup>-1</sup>, 563 kg body weight, 67 days in milk in average) according to a replicated 3 x 3 Latin square design during the following winter. Periods were 3 weeks long. Pre-experimental and post-experimental periods (2 weeks each) were conducted respectively on sorghum and maize silage diets to compare average intake of these silages with other better known silages. Cows were individually fed the experimental diets *ad libitum* each morning, using electronic Calan gates to control individual access to each manger. The amount of silage was determined to obtain 10% of daily refusals. The three diets of cows were balanced with the same amount and nature of concentrates (6 kg of energy rich commercial concentrate per cow per day). Offered forages were daily sampled and bulked per period. Fibre (Neutral Detergent Fibre, Acid Detergent Fibre and Acid Detergent Lignin), crude protein and *in vitro* dry matter digestibility (IVDMD) of the 3 silages were determined for each experimental period. Forage intake was recorded daily from d10 to d21 of each period. Milk composition (fat and protein contents) was measured on 6 consecutive milkings in each period. Data were submitted to analysis of variance.

## Results and discussion

The mean characteristics of the experiment are given in Table 1 (costs), Table 2 (crops), and Table 3 (animal performances).

The direct costs of the crops were low because of the lack of pesticides. Mixtures saved the costs of nitrogen fertilizer but their costs of seeds were higher.

Table 1. Cropping costs.

Treatments	Fertilizers (€ ha <sup>-1</sup> )	Seeds (€ ha <sup>-1</sup> )	Pesticides (€ ha <sup>-1</sup> )	Total (€ ha <sup>-1</sup> )
T1 triticale	62	88	0	150
T2 binary mixture	0	133	0	133
T3 complex mixture	0	132	0	132

The mixtures (T2 and T3, mean value) provided a lower forage yield (- 13%) than the pure triticale (T1). But the quality of the mixtures was also higher, in terms of the crude protein content (+ 25%), the NDF content (- 12%) and the digestibility (+ 10%). Among the two mixtures, T2 had a higher yield than T3 and a higher crude protein content.

The triticale crop was harvested too late and provided a high DM content (37.7%) compared to the mixtures (28.2 and 26.5% for T2 and T3). Cows fed the mixtures had a significantly higher intake than cows fed the triticale diet (+ 17%). This higher intake combined with the higher quality of the forage led to a higher milk production (+ 1.4 kg milk) but lower fat and protein concentrations (- 2 and - 0.7 points). The complex mixture (T3) had the highest intake (10.9 kg) and the better milk production (21.4 kg). The diminution of the proportion of triticale in the diet led to higher animal performances.

Table 2. Crops and forage characteristics.

Treatments	DM yield (t ha <sup>-1</sup> )	CP (g kg <sup>-1</sup> )	NDF (g kg <sup>-1</sup> )	ADF (g kg <sup>-1</sup> )	ADL (g kg <sup>-1</sup> )	IVDMD (g kg <sup>-1</sup> )
T1 triticale	8.9	79	638	374	37	470
T2 binary mixture	8.1	103	568	363	48	516
T3 complex mixture	7.4	94	566	360	45	518
significance	-	**	*	ns	**	*

Significance \*  $P < 0.05$ ; \*\*,  $P < 0.01$ .

DM: Dry matter; CP: Crude protein ; NDF: Neutral detergent fibre; ADF: Acid detergent fibre;  
ADL: Acid detergent lignin detergent fibre; IVDMD: *in vitro* dry matter digestibility.

Table 3. Intake and animal performances.

Treatments	Intake (kg d <sup>-1</sup> DM)	Dry matter (g kg <sup>-1</sup> )	Milk (kg d <sup>-1</sup> )	Milk fat (g kg <sup>-1</sup> )	Milk protein (g kg <sup>-1</sup> )
T1 triticale	9.0	377	19.6	41.8	27.8
T2 binary mixture	10.1	282	20.6	39.5	26.7
T3 complex mixture	10.9	265	21.4	40.0	27.4
significance	***	-	***	*	**

Significance \*  $P < 0.05$ ; \*\*,  $P < 0.01$ ; \*\*\*,  $P < 0.001$ .

## Conclusions

The results of the present study confirm that whole-plant cereals and mixtures are able to provide an acceptable biomass yield at a low cost. In this experience, opposite effects were observed between crop yields and animal performances. The presence of legumes improved silage quality and dairy cow performance. However, the digestibility and ingestibility of these silages were lower compared to typical values of maize or sorghum. Nevertheless in crops - forage rotations they can be useful for rearing ruminants without irrigation requirements and with low levels of inputs. Further investigations are to be conducted on the choice of species and cultivars (barbs occurrence, genetic variability), on the optimization of the crop (pesticides, fertilization), on the choice of the harvest date, on the silage particle size and on the complementation management.

## Acknowledgements

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# The effectiveness of grazing management in terms of milk production on sub-mountain dairy farms

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## Abstract

The aim of this study was to survey the seasonal changes in pasture herbage allowance, its chemical composition and the milk yield during the 2006 grazing season on two sub-mountain dairy farms in the Czech Republic. The examined pastures with *Lolio-Cynosurelion* vegetation were located at an altitude of 730 m a.s.l. (farm 1) and 575 m a.s.l. (farm 2). They were grazed in rotation by a herd of 95–122 cows and 1-ha representative areas were established in pastures. The daily milk yield decreased gradually during the grazing season. A significant depression of herbage nitrogen content and enhancement of acid detergent fibre content were recorded between June and subsequent summer months. A fodder supplement of higher nitrogen content (e.g. red clover) fed indoors is suggested as a means of improvement in the summer feed ration on the surveyed farms.

Keywords: pasture management, grazing, milk yield, ADF content, nitrogen content, cattle

## Introduction

In Central Europe the semi-natural grasslands form up to one-third of agricultural land (Zimková *et al.*, 2007). The predominant forms of utilization there are seasonal pasture (May–October) and grass harvested for hay or silage. Minimal sward restoration activities, in terms of resowing or artificial fertilization, are applied in pastures in the Czech Republic. The pasture management is confined to cutting and mulching once or twice in the growing season. Such farming can be viewed as environment-friendly, supporting the sward patchiness and habitat, flora and fauna diversity. The extensive utilization of grasslands is further underlined by a decline in cattle numbers over the last two decades, and has recently been reported as 58 cows per 100 ha of permanent grassland (Kvapilík *et al.*, 2007). In the sub-mountain dairy farms, higher individual milk yields have been recorded during the grazing season, in comparison with winter confinement (Frelich *et al.*, 2006). The aim of this study was to survey the coincidental changes in pasture herbage allowance, its chemical composition and the milk yield during the grazing season on sub-mountain farms.

## Materials and methods

Two dairy farms located in the foothills of Šumava Mountains in the Czech Republic (730 and 575 m a.s.l., farm 1 and farm 2, respectively) were selected for this study carried out in 2006. During the grazing period of May to October the herds consisted of 97–122 cows of Czech Pied breed on farm 1, and 95–101 cows of Czech Pied and Holstein breeds on farm 2. The half-a-day grazing (between morning and evening milking) was practised on farm 1. During milking the cows were offered mineral supplements and a grain concentrate (5 kg DM cow<sup>-1</sup> day<sup>-1</sup>). On farm 2 the cows also remained at pasture overnight, and during milking they were offered mineral supplements and, in addition, cows with a daily milk yield of > 12 litres were also fed a grain concentrate (0.4 kg DM day<sup>-1</sup> per litre of produced milk). From July a fresh cut of herbage was offered to cows during milking on both farms: on farm 1 a pasture sward (3 kg DM cow<sup>-1</sup> day<sup>-1</sup>) and on farm 2 herbage from a sown grass-legume-red clover



mixture (seed weight proportions of 46-46-8) of 4 kg herbage DM cow<sup>-1</sup> day<sup>-1</sup>. The vegetation of the pastures appertained to the *Lolio-Cynosurenia* suballiance. The pasture herbage was collected inside a one-hectare representative area before the beginning of the grazing, from either 3 or 2 plots of 10 m<sup>2</sup> area (farm 1 and 2, respectively). On farm 1, the samples were collected in May (no grazing), in June (9-days grazing), in August (9-days grazing) and in September (15-days grazing). The examined area was placed inside a 12 ha pasture field grazed in rotation with other fields. On farm 2, two pasture fields (field A, 42 ha; field B, 52 ha) were grazed in rotation. A representative area was established in both the fields and samples were collected successively: in May in field A (38-days grazing), in June in field B (18-days grazing), in July in field A (11-days grazing) and in July in field B (24-days grazing). The herbage was cut 3 cm above ground using cutting machine Husqvarna 323 R. Collected herbage was dried for 6 hours at 105°C and after subsequent weighing the dry herbage mass per 10 m<sup>2</sup> (DM) was determined. A single sample of herbage offered to cows indoors was collected in August and in September on farm 1, and in July on farm 2. The content of nitrogen was measured by Kjeldahl method. The acid detergent fibre (ADF) and neutral detergent fibre (NDF) were analysed using an *in-vitro* method (Van Soest and Wine, 1967). The average milk yield per cow was calculated on the basis of daily raw milk production of the herd and the number of cows in milk. Only the recording days when cows grazed on pastures with representative areas were considered in the milk yield calculation. One-way analysis of variance followed by Tukey's multiple comparison test (Statistica 6.0) were used to evaluate the differences in milk yields, DM, nitrogen, ADF and NDF contents between the grazing periods.

## Results and discussion

Table 1. Mean dry herbage mass (DM) from 10 m<sup>2</sup> plots inside pasture area (3 or 2 samples on farm 1 and 2, respectively), the mean herbage quality characteristics (nitrogen, acid detergent fibre (ADF) and neutral detergent fibre (NDF) contents in DM) and mean daily milk yields during pasture. Means with different letters within the same column are significantly different: \*  $P < 0.05$ ; \*\*  $P < 0.01$ ; \*\*\*  $P < 0.001$ ; ns is non significant ( $P > 0.05$ ). Cut herbage is fresh cut herbage fed indoors (single samples).

Period		DM (g)	Nitrogen (%)	ADF (%)	NDF (%)	Milk yield (kg)
		Mean	Mean	Mean	Mean	Mean
Farm 1	May	3154 a	21.9 a	24.1 a	41.5 a	-
	June	3820 a	15.6 a	27.2 a	46.9 a	15.9 a
	August	2471 b	13.6 b	35.7 b	59.5 b	15.0 ab
	September	1485 b	16.6 ab	30.4 c	52.6 b	14.9 b
		***	*	**	***	*
Cut herbage	August		14.6	42.4	56.9	
	September		16.3	41.4	53.7	
Farm 2	May, A	1007	19.4 a	24.7 a	40.1	20.8 a
	June, B	789	19.9 a	26.2 a	49.7	20.6 ab
	July, A	1395	13.7 b	30.3 b	46.9	20.1 b
	July, B	1666	15.2 ab	30.6 b	47.4	19.2 c
		ns	**	**	ns	***
Cut herbage	July		18.3	30.5	51.0	

The milk yields gradually decreased during the pasture season on both farms (Table 1). Due to year-round calving, the mean number of days in milk of cows was prolonged insignificantly between June and August (from 161 to 168 days on farm 1 and from 161 to 176 days on farm 2). The stage of lactation was unlikely to explain the decline in milk production. No significant change in DM allowance was found on farm 2, whereas on farm 1



the DM allowance was significantly lower in August than in June. Nevertheless, the amount of pasture herbage available, about 28.3 kg DM cow<sup>-1</sup> day<sup>-1</sup> in August (116 cows grazed 12 ha of pasture during 9 days) did not seem to limit DM intake. The cows were also supplied with herbage fed indoors. A significant change in nutritional quality of pasture was, however, found between June and July on both farms. The nitrogen content decreased, whereas ADF content (and NDF content on farm 2) increased in this period. Higher fibre content and lower nitrogen supply can depress milk yields in cows (Romney and Gill, 2000). The chemical composition of herbage offered indoors was similar to that of the pasture sward and could not have influenced substantially the nutritional quality of the feed ration. A supplementation of fodder of higher nitrogen and lower fibre content, red clover for example, might be a convenient way to improve the summer feed ration on farms such as those surveyed here. Red clover is readily accepted by cows, and it can enhance their nitrogen intake and hence the milk production.

## Conclusions

Gradual depression of milk yields during the pasture season was recorded on two submountain dairy farms. The depression of nitrogen content and enhancement of ADF content were found in representative areas of pastures between June and subsequent summer months. A fresh cut of fodder of higher nitrogen and lower fibre content is suggested as a means of improvement in the summer feed ration on the surveyed farms.

## Acknowledgement

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## Effect of grass silage chop length on chewing activity and digestibility

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### Abstract

Round bale grass silage harvested early (D-value 757 g kg<sup>-1</sup> DM) or at a normal (D-value 696 g kg<sup>-1</sup> DM) time was used to study the effect of harvesting time, chop length and their interaction on chewing activity and digestibility by dairy cows. Six early lactating Norwegian Red cows were used in a 6 x 6 Latin square with 3-week periods. Chewing activity was measured using IGER Behaviour recorders, and digestibility was measured by total collection of faeces. The two silages were fed long (170 mm), coarsely chopped (55 mm), or finely chopped (24 mm median particle length). Cows were fed silage *ad libitum* and supplemented with 6 kg concentrate. Early harvested silage significantly decreased total ration eating (ET), rumination (RT) and chewing time (CT) per kg silage DM compared with normal harvested silage (CT = 38 vs. 46 min kg<sup>-1</sup> DM). Chopping of silage reduced CT significantly, mainly due to reduced ET, CT = 45, 41 and 39 min kg<sup>-1</sup> DM for rations with long, coarsely and finely chopped silage, respectively. Grass silage chop length did not influence diet digestibility, but there was a significant effect of harvesting time on digestibility. No interaction between harvesting time and chop length was found, neither for chewing activity nor digestibility.

Keywords: silage, chop length, cows, chewing activity, digestibility

### Introduction

A reduction in forage particle size will usually increase intake of forage based rations, but is highly influenced by the forage: concentrate ratio. Lack of physical structure in dairy cow diets can result in decreased chewing activity, ruminal buffering and fibre digestion, and digestive disorders like rumen acidosis and parakeratosis may appear (Kaufman, 1976; De Brabander *et al.*, 1999; Tafaj *et al.*, 2007; Zebeli *et al.*, 2007). The objective of this study was to evaluate the effect of particle length of grass silage on chewing activity, rumen fermentation and whole tract digestibility.

### Materials and methods

Round bale grass silage harvested early on 1-2 June 2005 or at a normal harvesting time on 16 June was used to study the effect of chop length and harvesting time by chop length interaction on chewing activity and digestibility by lactating dairy cows. The silages were fed *ad libitum* to six Norwegian Red dairy cows (BW 618 ± 57 kg, 27 ± 9 d.i.m.) in second to sixth lactation in a 6 x 6 Latin square experiment with six three-week periods. The cows were supplemented with 6 kg d<sup>-1</sup> of concentrate divided in four equal meals. Information regarding ensiling and chopping procedures and chemical composition of herbage, silage and concentrate, as well as the animals, is given by Randby *et al.* (2008). Chewing activity for 24 hours was measured using IGER behaviour recorders (Rutter *et al.*, 1997) simultaneously for the six cows during week two in each of the experimental periods. Digestibility was measured by total collection of faeces for three subsequent days in week three.

## Results

The silage median particle length measured by hand sorting is given in Table 1. Early harvesting and fine chopping increased silage intake during the chewing measurements (Table 1), in line with observations from the whole experimental periods (Randby *et al.*, 2008). There was no significant difference in eating time (ET) in minutes between the two harvesting times (Table 2), but early harvested silage showed a significant shorter total ruminating (RT) and chewing time (CT) compared with normal harvested silage (RT = 508 vs. 573 min; CT = 871 vs. 960 min). Early harvested silage significantly decreased eating, ruminating and chewing time per kg DM (CT = 39 vs. 46 min kg<sup>-1</sup> DM), but not per kg NDF (CT = 99 and 100 min kg<sup>-1</sup> NDF). Eating time tended to be shorter for finely and coarsely chopped silage compared with long silage (ET = 359, 358 and 409 min, respectively), but chopping did not significantly affect ruminating or total chewing time. Although total chewing time in minutes was unaffected by silage chop length, the time consumption per kg DM or NDF was significantly reduced by chopping, mainly due to reduced ET (CT = 45, 41 and 39 min kg<sup>-1</sup> DM for rations with long, coarsely and finely chopped silage, respectively). Silage chop length did not significantly affect rumen fermentation parameters (data not shown) or total ration digestibility (Table 2).

Table 1. Median particle length (MPL) of silages and feed intake of lactating dairy cows.

	Early harvested silage			Normal harvested silage		
	Long	Coarse	Fine	Long	Coarse	Fine
MPL, mm	184	57	24	153	53	23
Silage, kg DM	16.44	16.55	18.59	14.47	15.83	16.71
Concentrate, kg DM	5.24	5.23	5.22	5.27	5.27	5.27
Total ration, kg DM	21.68	21.78	23.81	19.74	21.10	21.98
Silage, kg NDF	7.03	6.95	7.96	7.71	8.45	8.92
Total ration, kg NDF	8.22	8.14	9.14	8.91	9.65	10.12
Silage NDF, g kg <sup>-1</sup> BW	11.7	11.2	12.8	12.5	13.9	14.4

Table 2. Effect of harvesting time and grass silage chop length on chewing activity and total ration digestibility of lactating dairy cows.

	Early harvested silage			Normal harvested silage			SEM <sup>1</sup>	Effect (P)		
	Long	Coarse	Fine	Long	Coarse	Fine		Time <sup>2</sup>	Chop <sup>3</sup>	T x C <sup>4</sup>
<i>Eating time</i>										
Min day <sup>-1</sup>	383	363	345	435	353	372	21	ns <sup>5</sup>	0.08	ns
Min kg <sup>-1</sup> DM	17	16	14	22	17	17	1.1	*	*	ns
Min kg <sup>-1</sup> NDF	45	42	37	48	37	37	2.6	ns	**	ns
<i>Ruminating time</i>										
Min day <sup>-1</sup>	504	509	510	555	579	584	14	***	ns	ns
Min kg <sup>-1</sup> DM	23	23	21	28	27	26	0.8	***	ns	ns
Min kg <sup>-1</sup> NDF	60	60	54	62	60	57	2.0	ns	0.09	ns
Min kg <sup>-1</sup> NDF silage	70	70	62	71	69	65	3	ns	0.07	ns
<i>Total chewing time</i>										
Min day <sup>-1</sup>	887	872	855	990	932	956	17	***	ns	ns
Min kg <sup>-1</sup> DM	40	39	35	50	44	43	1.1	***	**	ns
Min kg <sup>-1</sup> NDF	105	102	91	110	97	94	3.0	ns	**	ns
<i>Digestibility</i>										
Dry Matter	.794	.788	.791	.730	.733	.735	.008	***	ns	ns
Organic matter	.804	.799	.803	.739	.742	.744	.008	***	ns	ns
Crude protein	.754	.750	.757	.709	.712	.713	.010	***	ns	ns
NDF	.772	.769	.773	.690	.693	.696	.010	***	ns	ns

<sup>1</sup>SEM = Standard error of the mean, <sup>2</sup>Harvesting time, <sup>3</sup>Chop length, <sup>4</sup>Harvesting time x chop length interaction,

<sup>5</sup>Statistical significance: \*  $P < 0.05$ ; \*\*  $P < 0.01$ ; \*\*\*  $P < 0.001$ ; ns =  $P \geq 0.1$ .

## Discussion

Decreasing the forage particle length in grass silage-based TMR diets from 25 to 10 mm has recently been shown to reduce both the total chewing time in minutes and chewing time per kg DM, of early lactating cows by approximately 20% (CT = 900 vs. 725 min d<sup>-1</sup>, 41 vs. 33 min kg<sup>-1</sup> DM), but still forage NDF concentration was a much better determinant than particle size for DM intake, rumen fermentation characteristics and diet digestibility (Tafaj *et al.*, 2007) in line with the present study. A study of Zebeli *et al.* (2007) showed that altering the forage particle length from 6 to 30 mm of hay (566 g NDF kg<sup>-1</sup> DM) in a low concentrate diet (0.21) significantly increased the chewing time (719 vs. 816 min d<sup>-1</sup>) without affecting ruminal fermentation or passage rate. They concluded that the response of chewing or ruminating activity alone seemed to be insufficient to assess the dietary physical effectiveness or fibre adequacy in limit-fed dairy cows when high-concentrate diets were fed separately. Chopping of grass silage down to 24 mm MPL in the present study did not affect digestibility, but Tafaj *et al.* (2007) showed a negative effect on NDF digestibility when forage particle length was reduced from 25 to 10 mm (0.55 vs. 0.49). This supports the view that approximately 20 mm particle length may be an optimal length in dairy rations (De Brabander *et al.*, 1999). The maximum possible chewing time of 1000 min per day considered by Mertens (1997) was nearly achieved for long silage at normal harvesting time in the present study. The mean chewing time per kg DM ingested was 35-50 min kg<sup>-1</sup> DM, which is well above the minimum recommendation of Sudweeks *et al.* (1981), of 30 min kg<sup>-1</sup> DM intake for limiting the risk of digestive disorders.

## Conclusions

Chopping of silage increased feed intake, but did not significantly affect the eating, ruminating or total chewing time. However, chopping did decrease eating and total chewing time per kg DM and per kg NDF of silage based rations. Chop length of silage had no effect on rumen fermentation or whole tract digestibility of DM, OM, CP or NDF in the present diets. No interaction between harvesting time and chop length was found, neither for chewing activity nor digestibility. No indications of lack of physical structure were detected when very early harvested grass silage was chopped to 24 mm particle length and fed *ad libitum* to dairy cows.

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# The effects of grazing pressure and concentrate use on efficient dairy production in Galicia

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## Abstract

Stocking rate is a very important factor for pasture management and utilization when dairy production is based on farm resources. Two experiments were carried out during two years to study optimal stocking rate and concentrate supplementation of grazing dairy cows during spring. In the first year, three treatments were compared: 4 kg cow<sup>-1</sup> of concentrate with a stocking rate of 3.3 cows ha<sup>-1</sup> (LL), and 8 kg cow<sup>-1</sup> with two stocking rates 3.7 (HL) and 5.3 cows ha<sup>-1</sup> (HH). Dairy milk yields were 22.8 kg cow<sup>-1</sup> for low concentrate and 28.9 and 28.5 kg cow<sup>-1</sup> for the treatment with a high concentrate level at the lower and higher stocking rates, respectively. In the second year two treatments were compared: 4 kg cow<sup>-1</sup> of concentrate with a stocking rate of 4.3 (LL) and 5.6 (LH) cows ha<sup>-1</sup>. No significant differences in milk production were found, with a mean milk yield of 24.6 and 25.5 kg cow<sup>-1</sup> at the low and high stocking rate, respectively. The results show that under these grazing conditions, it may be possible to increase the stocking rate without decreasing milk quality and production per cow. Special care in grazing management is essential under these conditions.

Keywords: farm resources, stocking rate, pasture management

## Introduction

Milk production systems must be competitive and sustainable. This is possible in the humid north west of Spain by reducing costs of production through maximizing the farm's own resources, using minimum rates of concentrates.

A question arises about the limits of grassland production, and how this can be transformed economically into animal production. Grazed grass should be the main source of nutrients, and individual cow performance requires a high dry matter (DM) intake and implies a pasture management that maintains herbage on offer, while at the same time good pasture utilization is needed for the best pasture quality. Increasing the grazing pressure as a consequence of reducing concentrate inputs would have the beneficial effect of improving both quality and intake of the grass.

Farmers need to predict grass growth and herbage intake of dairy cows, and consequently this should enable increased confidence in, and reliance on, grazed grass for milk production. Tools to provide information on grazing management for dairy cows have been developed (Mayne *et al.*, 2004) and more trials on the stocking capacity of grassland in our conditions are needed when concentrate is limited to critical moments of lactation.

The objective of this paper was to compare the effect of different stocking rates and concentrate supplementation on milk and pasture production and quality during spring in Galicia (NW Spain).

## Materials and methods

A grazing experiment was made during the spring of two years. In the first (2005), 54 Friesian cows calving around mid-February, received 5 kg of concentrates (132 g kg<sup>-1</sup> crude protein, CP) and 7 kg cow<sup>-1</sup> of maize silage, (348 g kg<sup>-1</sup> DM and 95 g kg<sup>-1</sup> CP), for 60 days, yielding 29.5 kg cow<sup>-1</sup> of milk. In early April the cows were divided into three groups grazing different



areas with the same herbage on offer, around 20 cm high, and the same post-grazing height, 5 cm, during five rotations in spring, of a perennial ryegrass-white clover sward. In the second year (2007), 44 Friesian cows, calving mid-February, grazed in 2 groups and received 4 kg cow<sup>-1</sup> day<sup>-1</sup> of concentrate, using different areas with 5 and 6 rotations in spring (32 and 26 days) respectively, to give two grazing pressures. Treatments of both years are in Table 1.

Table 1. Experimental groups at different grazing pressures during the spring of two years.

Treatments		Concentrate (kg cow <sup>-1</sup> )		Stocking rate (cow ha <sup>-1</sup> )	
Conc. level-stocking rate		1 <sup>st</sup> year	2 <sup>nd</sup> year	1 <sup>st</sup> year	2 <sup>nd</sup> year
LL	Low-low	4	4	3.3	4.3
HL	High-low	8		3.7	
HH	High-high	8		5.3	
LH	Low-high		4		5.6

It was necessary to adjust the grazing pressure in both years to achieve the best utilization of the grassland. High DM intake goes with high herbage on offer; at the same time the ration had to be with the best pasture quality. Increasing instant grazing pressure seems to maintain quality and meet the needs of the productive herd (Gonzalez *et al.*, 2007).

Daily milk yields per cow were recorded through the whole lactation, and weekly samples were analysed for milk parameters. Pasture DM intake was estimated as the difference between pre- and post- grazing herbage mass, using five quadrats (0.5 m<sup>2</sup>) per paddock, and botanical composition was determined by hand separation, and sampled for dry matter content. Samples were sent to the laboratory to determine crude protein, fibre contents and organic matter digestibility by NIRS.

## Results and discussion

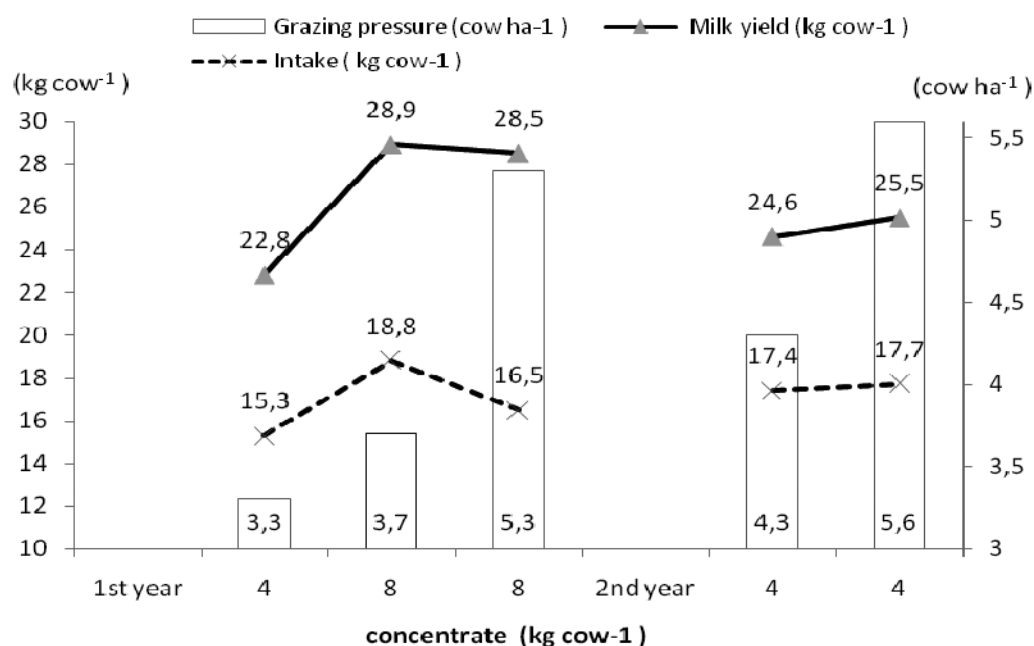


Figure 1. Milk yield (solid line), DM Intake (dotted line) and average stoking rate (bars) from dairy cows at two levels of concentrate, two grazing pressures and two years.

Figure 1 shows milk yield, total DM intake and stocking rate obtained in the experiments. The measured herbage DM intake during the first spring in the three herds was 11.3, 10.8 and 8.5 kg cow<sup>-1</sup>, respectively. In the spring of the second year the grazing pressures were not as different as in the first year, for the two groups. The DM herbage on offer was *ca.* 17 kg cow<sup>-1</sup> and the DM intake of LL and LH treatments, 13.4 and 13.7 kg cow<sup>-1</sup>, had not significant differences. Total milk yield during the experiment in spring was 3800 and 4100 kg cow<sup>-1</sup>.

Annual results from the first year were summarized in a previous paper (Gonzalez *et al.*, 2007) and milk yield reached 8800 kg cow<sup>-1</sup> (Gonzalez *et al.*, 2007). A total of 1430, 2260 and 2560 kg cow<sup>-1</sup> of concentrate were used annually, meaning levels of 200, 260, 290 g kg<sup>-1</sup> per treatment, considerably less than the levels of 450 g kg<sup>-1</sup> of milk produced, as used in the intensive dairy farms in the region (Barbeyto, 1999).

A better utilization of the pasture was reached with the groups with higher grazing pressure, with a tendency towards better quality of grass. The average pasture CP was 135, 140 and 161 g kg<sup>-1</sup> for groups LL, HL and HH, respectively, in the first year, but differences among groups were not significant. The evolution of CP in spring, 176, 116 and 87 g kg<sup>-1</sup> in April, May and June, shows a drop in quality, due to lower pasture utilization in the reproductive stage of the grass. Other parameters of pasture quality were similar in the three independent grazing areas, 913 g kg<sup>-1</sup> OM, 813 g kg<sup>-1</sup> OMD, 290 g kg<sup>-1</sup> ADF, 483 g kg<sup>-1</sup> NDF, and 192 g kg<sup>-1</sup> of carbohydrates as WSC. Sward characteristics, due to an increase in stocking density when feeding less concentrate, should be regarded as of equal importance to the pre- and post-grazing sward heights in providing adequate herbage on offer (Peyraud and Gonzalez, 2000). The effect of a 29% reduction of the grazing area in spring, group HH in relation to group HL in the first year, brought an increase of a 41% in total milk yield per ha (from 12,800 to 18,000 kg). In the second year the grazing area was reduced by 20%, and it was possible to produce 36% more milk per ha (from 16,600 to 22,600 kg).

## Conclusions

The results show that stocking rate can be increased when using concentrate at spring grazing, without decreasing milk production or quality. Stocking rate can be as high as 5.6 cows ha<sup>-1</sup> during spring in Galician conditions. It is possible to reach milk yields of 8800 kg cow<sup>-1</sup> with only 290 g per litre of concentrate, considerably less than the 450 g. per litre used on intensive farms. Increasing grazing pressure can produce around a 40% gain in milk yield per ha. Special care on pasture management is needed for high animal intake when using high levels of herbage on offer. Increasing grazing pressure in this situation could bring better pasture utilization and higher pasture quality.

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## Horse grazing: practices in Franche-Comté region of France

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### Abstract

Franche-Comté is one of the major regions in France for workhorse breeding. In this hill country characterized by grassland landscapes, horses are widely owned by farmers. Most of the farms specialize in cattle milk production according to specifications in order to make cheese with a registered designation of origin (Comté). Horse owners have usually used mixed grazing by cattle and horses. In order to identify their practices, we interviewed eight farmers about the way they managed horse grazing. Four types of management were found: horse grazing was simultaneous (mixed grazing), alternate, either the beginning or the end of the grass growth season, and winter grazing. The farms were divided into three groups taking account of (i) the characteristics of the farms and (ii) the importance of horse production in the farming system. Within each farm group, not all types of horse grazing management were used by all of the farmers. The results point out different aspects that could be studied in more detail.

Keywords: mixed grazing, horses, farming system, permanent grassland

### Introduction

Grazing is often used as a management tool for grassland maintenance and the different herbivore species, grazing alone or in combination, are considered to be complementary regarding grazing management, parasite control and animal feeding (Lendal *et al.*, 1998; Loucougaray *et al.*, 2002). Among the different species, horses seemed to take a singular place (Fleurance *et al.*, 2007). In France, Franche-Comté is one of the major regions for workhorse breeding: the number of brood mares reaches 8.1% of the French herd (Agreste, 2002; Lemaire, 2003). In this hill country characterized by grassland landscapes, horses belonged mostly to farmers. Most of the farms specialized in cattle milk production with respect to specifications in order to make cheese with a registered designation of origin (Comté). New legislation has integrated horses for stocking rate calculation. This has led to higher stocking rate on farms which received subsidies for low intensity herbage management and to a trend towards the disappearance of horse herds on dairy farms. In order to save workhorse breeding in Franche-Comté, the National Association of Comtois Workhorses was interested in identifying the advantages of mixed grazing. In Franche-Comté, farmers usually used mixed grazing by cattle and horses but their practices were poorly known. A study has been carried out in order to characterize farmers' practices for horse grazing management.

### Materials and methods

We described grazing management by surveys performed on 8 farms, chosen in order to take into account the trend of horse production (meat or leisure). In these farms, the number of horses was at least five. We devised the questionnaire to evaluate in detail grazing management: (i) the organization of grazing for all the paddocks on each farm, (ii) the kind and number of grazing animals, the grazing duration for each paddock. To collect data on

explicative factors, the farmers were also interviewed about (i) farm structure (total area and labour force, field pattern) (ii) production system and (iii) advantages of horse grazing.

## Results

In the interviewed farms, the Comté workhorse breeding system was similar to those described by Micol et al. (1997) in other regions. The foaling mainly took place in March and April; the contribution of grasslands to the total diet was high during the lactation period. After weaning, usually in October, brood mares kept on grazing before wintering from January to March. During the winter period, brood mares and young horses received hay and cereals. The foals bred for meat (3 farms) were sold when they were one year old.

We used the importance of horse production in the farming system as a typological key to sort the farms. Three groups were identified. In the first G1 (3 farms), the horse unit was a complement to cattle milk production: two farms produced milk with a registered design of origin (stocking rate  $< 0.8$  LU (livestock unit)  $\text{ha}^{-1}$ ); one fed dairy cows with maize silage and produced standard milk (stocking rate was *ca.*  $2$  LU  $\text{ha}^{-1}$ ). The ratio of cattle-to-horse was 1 brood mare for 5 to 7 dairy cows. Farmers produced various horse products (meat and/or leisure). In the second G2, the two farms were also dairy farms (Comté milk); they had the same number of dairy cows and brood mares or stallions. The stocking rate was *ca.*  $1.2$  LU  $\text{ha}^{-1}$ . Bred horses were intended for reproduction or leisure. In the last group G3 (3 farms with the smallest farm area, less than 40 ha), there was no cattle herd (or only a few cows). Farmers bred horses (20 to 50) first for leisure, then for meat when they had a high number of animals. They simultaneously developed another activity: farm-based tourism, vegetable cropping or a non-agricultural job. The proportion of grasslands cut was higher for G1 and G2 than for G3, respectively more than 50% and less than 50%, but farmers of G2 were not self-sufficient for forage and bought hay for winter feeding.

Farmers found that horse grazing benefited grasslands and it was of major interest to improve the use of grass production and the botanical composition of grasslands. All data on grazed paddocks were analysed to identify the different ways horse grazing was implemented. Four types of management were found. The first one consisted of an alternate grazing implemented at the beginning of the grass growth period: horses first grazed paddocks which would be then assigned to dairy cows. A second type of alternate grazing was implemented: at the end of the grazing period (September, October), the kind of grazing animal changed. Cattle first grazing at the beginning of the pasture season were replaced by horses (alternate grazing CH). The reverse situation was also found, horses first then cattle (alternate grazing HC). The third one was a mixed grazing: heifers or dry cows grazed simultaneously with horses, mainly suckled mares. The last one was a winter grazing using horses alone, mares, whose foals were weaned, in November and December or young horses in January and February.

Considering the farm scale, the frequency and diversity of these types of management depended on the group that the farm belonged to (Table 1).

Table 1. Practice frequency for horse grazing management taking into account farm group.

Management used in group by: all the farmers (++) ; only some farmers (+); none (0)

C: Cattle. H: Horses.

Horse grazing management	G1	G2	G3
Alternate grazing - beginning of the season	++	0	0
Alternate grazing - end of the season	+	0	+
	CH		HC & CH
Mixed grazing	++	++	+
ratio horse/cattle (paddock scale)	1/3	1/2	2/1
Winter grazing	++	++	+

The farmers of G1 implemented the highest diversity and practice frequency for horse grazing management. Only one farmer of G1 did not use alternate grazing at the end of the season. In G2, two types of management were used: the diversity was the lowest. The practice frequency of each management was low in G3: there were various types of management but not on all farms. The farmers of this group were used to exchanging animals (horses and heifers) with their neighbours. They also used horses for topping of grasslands cut for hay.

## Discussion and conclusions

Farming systems and types of horse grazing implementation seem to be linked, whereas no statistical evaluation is possible due to the low number of farmers interviewed. However, the results point out different aspects that could be studied in more detail.

Farmers consider that alternate grazing and winter grazing tends to improve botanical composition of swards. Winter grazing is the most frequent management used by the interviewed farmers, particularly on grasslands previously cut for hay. Alternate grazing at the beginning of the season is only found in G1 in order to give better herbage to dairy cows. Alternate grazing at the end of the season is not used by all farmers. Furthermore, the order of grazing species differs between farms. This point has been recognized as an influence on the improving effect of horse grazing (Loiseau and Martin-Rosset, 1988). The real impacts on the improvement of botanical composition of these types of management are still to be quantified in further studies.

Most farmers use mixed grazing (i) to improve the use of herbage resources in low intensity management and to decrease horse breeding costs (G1), (ii) to eat herbage refused by one species and to decrease grassland maintenance costs (G2 and G3). Consequently, the ratio of cattle-to-horse grazing together differs between groups. This ratio has an impact on horse growth performances (Martin-Rosset *et al.*, 1984 in Micol *et al.*, 1997). However, this needs to be further evaluated in the local conditions of Franche Comté.

The interest of horse grazing is based on feeding behaviour which differs between horses and cattle: horses have a higher intake rate and can eat low quality forage (Martin-Rosset *et al.*, 1981; Hoskin and Gee, 2004; Fleurance and Duncan, 2005). It enhances the role of horse owners as land managers. Horse grazing implementation can take many forms as seen in the surveys. This diversity is a means to promote the maintenance of horse herds on farms of Franche-Comté.

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# Voluntary intake of silage in small and large ruminants depending on chemical composition and *in-vitro* gas production characteristics

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## Abstract

In order to better understand the regulation of silage dry matter intake (SDMI), the results of three recent Scandinavian feeding experiments, including 48 silages fed to steers, lambs or dairy cows, were compiled. The silages had been characterized by chemical analysis and an *in vitro* gas production (GP) recording technique with end point measurements of substrate residues. The relationship between feed variables and SDMI were investigated by simple linear and multiple regressions. Interesting linear relationships were found between GP parameters, chemical components and the voluntary SDMI. The best multiple regression relationship was found to be: relative daily SDMI (kg per 100 kg live weight) =  $-0.562 - 0.574 * (\text{Experiment}) + 0.00481 * \text{NDFD} - 0.411 * C$  ( $R^2 = 0.80$ , S.D. = 0.15). Experiment is the variable for data related to dairy cows (1 or 0), NDFD is the degradability of the neutral detergent fibre and C is the variable that regulates the switching characteristics of the GP profile. It was concluded that the GP technique is a powerful tool to describe silages from northern regions and the results may be valuable for predicting SDMI.

Keywords: silage, ruminants, chemical composition, in vitro characteristics, voluntary intake

## Introduction

In Scandinavia silage is commonly fed to dairy cows, growing cattle and sheep. It is therefore interesting to interpret data on silage dry matter intake (SDMI) including intake by different types of domestic ruminants. Due to climatic conditions, botanical composition and crop management, there is a considerable variation in silage quality which will have an impact on SDMI. Various quality parameters therefore have been used to predict SDMI (e.g. Huhtanen *et al.*, 2007). In high quality silages a large part of the degradable matter is soluble and readily degradable in the rumen. The gas production (GP) technique records the fermentation of both soluble and non-soluble components, which makes it interesting for the characterization of silages. The SDMI have also been explained by level of production and amount of concentrates (supplementation) in the diet (reviewed by Huhtanen *et al.*, 2007). From a practical perspective it is more valuable to predict the relative SDMI prior to feeding, without information on animal performance and diet composition. The objectives of this study was to investigate the possibility to develop models for prediction of the relative SDMI in small and large ruminants fed temperate silages, using information on the chemical composition and *in vitro* GP characteristics of the silages.

## Materials and methods

The data in the study were compiled from three recent studies on the relationships between the voluntary SDMI and the chemical and *in vitro* characteristics of 48 silages from northern regions in Scandinavia (Table 1). The silages were fed *ad libitum* to either steers (n = 24) (Krizsan and Randby, 2007), lambs (n = 9) (Hetta *et al.*, 2007b) or dairy cows (n = 15) (Hetta

*et al.*, 2007a). The silages were harvested from leys that consisted mainly of timothy (*Phleum pratense*), and meadow fescue (*Festuca pratense*) but some silages also contained red clover (*Trifolium pratense*), white clover (*Trifolium repens*) and chicory (*Cichorium intybus*). The feeds were characterized by chemical analysis and by an automated GP recording technique. The technique recorded the GP *in vitro* for 72 h when the feeds were suspended in buffered rumen fluid, thereafter the potential degradability of organic matter (OMD) and NDF (NDFD) was determined by filtration of the residues (Hetta *et al.*, 2007a). The cumulative GP data were fitted to a Michaelis-Mentens equation including three parameters: A (ml gas g<sup>-1</sup> OM) represents the asymptotic GP; B (h) is the time at which half of the gas (A) has been produced and C is a dimensionless parameter that determines the sharpness of the switching characteristics of the GP curve. The relationships between feed parameters and voluntary SDMI were explored with linear regression analysis. The experimental unit was the treatment means for each silage (n = 48). Multiple linear relationships were selected using the stepwise routine (MINITAB™ ver. 14.1) with  $\alpha$  to enter and to remove set to 0.15. Models were evaluated using the R<sup>2</sup> value and the fitted standard deviation (SD). The stepwise routines were performed with experiment (e.g. dairy cow, steer and lamb) as random variables.

Table 1. Description of the data sets used.

	SDMI	OMD	NDFD	A	B	C	DM	CP	NDF	WSC	LA	AcA	pH	NH <sub>4</sub> N
DairyCows														
Mean (15)	1.89	876	799	244	10.2	1.99	260	137	600	19	78	18	3.92	81
STDV	0.12	20	32	15	0.5	0.27	35	18	35	16	24	6	0.15	18
Min	1.66	846	731	217	9.4	1.52	201	100	533	0	24	7	3.75	51
Max	2.02	919	854	260	10.9	2.35	341	161	678	47	110	26	4.32	113
Lambs														
Mean (9)	2.52	896	817	251	8.7	2.07	273	134	501	17	82	17	3.97	85
STDV	0.34	29	45	26	0.9	0.11	49	20	52	16	22	5	0.15	11
Min	2.05	858	759	210	7.7	1.96	232	100	452	3	31	8	3.84	65
Max	3.09	946	894	296	10.3	2.3	386	172	587	46	101	25	4.25	97
Steers														
Mean (24)	2.38	879	805	228	8.9	2.26	213	174	544	33	49	29	4.56	142
STDV	0.19	10	8	16	0.5	0.23	18	9	33	15	24	15	0.33	50
Min	1.79	862	792	199	8.0	1.84	166	158	476	16	2.2	12	4.01	62
Max	2.65	895	823	264	10.2	2.79	237	193	601	71	102	65	5.26	255

Figures within brackets = n, SDMI = Silage Dry Matter Intake (kg DM 100 kg live weight<sup>-1</sup> day<sup>-1</sup>), OMD = Organic matter degradability (g kg<sup>-1</sup> OM), NDFD = Neutral detergent fibre degradability (g kg<sup>-1</sup> NDF), A = Asymptotic gas production (ml gas g OM<sup>-1</sup>), B = the time (h) when ½ of A is produced, C regulates the switching characteristics of the GP profile, DM = dry matter (g kg<sup>-1</sup> feed), CP = crude protein (g kg<sup>-1</sup> DM), NDF = Neutral detergent fibre (g kg<sup>-1</sup> DM), WSC = water soluble carbohydrates (g kg<sup>-1</sup> DM), LA = Lactic Acid (g kg<sup>-1</sup> DM), AcA = Acetic Acid (g kg<sup>-1</sup> DM) and NH<sub>4</sub>-N = AmmoniaN (g kg<sup>-1</sup> N).

## Results and discussion

Correlation coefficients between SDMI and feed parameters are presented in Table 2. All of the correlations are in agreement with the large study of Huhtanen *et al.* (2007), displaying the effects of fibre content and silage fermentation on SDMI. The results showed that the GP profiles are related to silage quality and SDMI. The best relationship from the multiple regression analysis in this study was: SDMI (kg 100 kg<sup>-1</sup> live weight day<sup>-1</sup>) = -0.562 - 0.574 \* (Experiment) + 0.00481 \* NDFD - 0.411 \* C (R<sup>2</sup> = 0.80, S.D. = 0.15, n = 48). Experiment is the variable for data related to dairy cows (1 or 0). Concentrates were only included in the diets of the dairy cows and the negative effect of 'dairy cow' in the model is more likely to be a substitution effect than a reduced digestive capacity. From the results one



can assume that silages that will promote intake should have a highly degradable fibre fraction (NDFD) and a rapid fermentation in the rumen.

Table 2. Correlations between SDMI and feed parameters.

	SDMI	OMD	NDFD	A	B	C	DM	CP	NDF	WSC	LA	AcA	pH
OMD	0.49												
NDF	0.50	0.87											
A	0.17	0.27	0.32										
B	-0.73	-0.57	-0.43	0.01									
C	0.04	0.17	0.10	-0.65	-0.31								
DM	-0.15	0.08	0.07	0.47	0.32	-0.56							
CP	-0.04	0.19	0.25	-0.45	-0.41	0.60	-0.68						
NDF	-0.59	-0.54	-0.26	0.08	0.73	-0.25	0.21	-0.26					
WSC	0.20	0.06	0.03	0.05	-0.23	-0.09	0.15	0.16	-0.15				
LA	-0.15	0.27	0.04	0.33	-0.13	-0.32	0.07	-0.45	-0.19	-0.39			
AcA	-0.04	-0.14	-0.05	-0.51	-0.05	0.67	-0.64	0.53	-0.08	-0.29	-0.35		
pH	0.33	-0.20	-0.01	-0.40	-0.16	0.44	-0.40	0.71	0.00	0.20	-0.83	0.50	
NH <sub>4</sub> N	0.11	-0.23	-0.14	-0.42	-0.15	0.56	-0.54	0.63	-0.05	-0.05	-0.59	0.61	0.86

Correlations > 0.29 are statistically significant ( $P \leq 0.05$ ). For abbreviations and units see Table 1.

## Conclusions

The GP technique is a powerful tool to describe the quality of high quality silages and the relative voluntary SDMI may be predicted from models based on GP parameters.

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# Faecal particle-size distribution from ewes fed grass silages harvested at different stages of maturity

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## Abstract

The aim of this experiment was to study the effect of maturity stage of grass at harvest on particle size in faeces from ewes fed grass silage *ad libitum*. Eighteen pregnant Swedish ewes bearing two foetuses were given one of three treatments as their only feed. The treatments were early (ECS), medium (MCS) or late (LCS) cut grass silage. The ECS, MCS and LCS silages contained 449, 578, and 634 g NDF kg<sup>-1</sup> and 166, 111 and 81 g crude protein kg<sup>-1</sup> DM, respectively. The *in situ* rumen indigestible NDF (INDF) was 77, 164 and 268 g kg<sup>-1</sup> of NDF; the degradation rate of digestible NDF (k<sub>d</sub>DNDF) was 64, 47 and 44 g kg<sup>-1</sup> h<sup>-1</sup> and DM intake was 2.5, 2.1 and 1.5 kg d<sup>-1</sup> for ECS, MCS and LCS silage, respectively. Faeces samples were collected during four days, washed in nylon bags, freeze dried and sieved into six sieving fractions; bottom bowl (B), 0.106 (C), 0.212 (D), 0.5 (S), 1.0 (M) and 2.36 (O) mm pore size. The proportions of particles in the B, C, D, S and O fractions were affected by cutting time of the silage ( $P < 0.003$ ). In conclusion, stage of maturity at harvest strongly affects the distribution of particle size in faeces from ewes fed grass silages.

Keywords: particle length, particle area, most frequent length, sieving technique, sheep

## Introduction

The size of feed particles varies among plant species and stage of maturity at harvest (Mertens, 1997). The particle size of forage and faeces has been measured by use of both dry and wet-sieving techniques (Nørgaard, 2006). The objective of the present research was to study the effect of stage of maturity at harvest of grass silage on particle-size distribution in faeces from pregnant ewes.

## Materials and methods

This experiment was conducted at Götala Research Station, Skara, Sweden in 2006. During the experiment, eighteen pregnant Swedish Finull × Dorset twin-bearing ewes, which were kept in individual pens, were fed three forage treatments *ad libitum*. The treatments were early, medium and late cut unchopped grass silages of a mixture of timothy (*Phleum pratense* L.), meadow fescue (*Festuca pratensis* L.) and perennial ryegrass (*Lolium perenne* L.), which were fed as sole feeds. The early, medium and late cut grasses were harvested from the same field on the 2nd, 12th and 21st of June during the spring growth cycle, respectively. The trial, which was conducted during two weeks as a part of an experiment that was performed on pregnant and lactating ewes, started one month before parturition (Nadeau and Arnesson, 2008). Forage intake was measured daily and 50 g faeces were collected twice a day during four days. Dry-matter (DM) contents in the silages and refusals were measured after drying at 60°C and in faeces after drying at 105°C to a constant weight. Chemical composition of the silages was analysed by conventional methods. Indigestible NDF (INDF) concentrations of



the silages were measured after a 288-h *in situ* incubation. Rates of degradation for digestible NDF ( $k_d\text{DNDF}$ ) in the silages were measured *in sacco* using 0, 2, 4, 8, 16, 24, 48, 96 and 168 h of incubation (Hvelplund and Weisbjerg, 2000). For wet sieving, 50 g faeces were soaked and rinsed with water on a 2.36-mm sieve and the number of particles longer than 10 mm (LP) was counted. For dry sieving, 15 g faeces were washed in nylon bags and freeze dried before being vertically shaken into six fractions during four minutes at an amplitude value of 1.5g by use of Retsch AS 200 shaking equipment. The six fractions included 5 sieves with square holes of 2.36 (O), 1.0 (M), 0.5 (S), 0.212 (D), 0.106 (C) mm and a bottom bowl (Nørgaard *et al.*, 2004). The arithmetic mean particle size (APS), the geometric mean particle size (GPS) and the most frequent particle size (Mode\_PS) were estimated according to Nørgaard (2006). Data on intake and faecal characteristics were analyzed statistically by using the Proc Mixed procedure of SAS (ver. 9.01, 2002).

## Results and discussion

Maturation of the grass increased contents of NDF, ADF, ADL and INDF but decreased the crude protein content (Table 1).

Table 1. Chemical composition and degradation characteristics of early cut (ECS), medium cut (MCS) and late cut (LCS) grass silages.

	ECS	MCS	LCS
DM, g kg <sup>-1</sup>	579 ± 6.7	661 ± 6.6	536 ± 25
Crude protein, g kg <sup>-1</sup> DM	166 ± 0.5	111 ± 0.5	81 ± 5.1
Ash, g kg <sup>-1</sup> DM	62 ± 0.2	65 ± 0.1	54 ± 0.3
NDF <sup>1</sup> , g kg <sup>-1</sup> DM	449 ± 1.1	578 ± 8.7	634 ± 1.6
ADF <sup>2</sup> , g kg <sup>-1</sup> DM	267 ± 1.4	357 ± 2.1	383 ± 6.3
ADL <sup>3</sup> , g kg <sup>-1</sup> DM	19 ± 0.1	39 ± 1.5	52 ± 0.7
INDF <sup>4</sup> , g kg <sup>-1</sup> NDF	77 ± 0.2	164 ± 0.2	268 ± 0.3
$k_d\text{DNDF}$ <sup>5</sup> , g kg <sup>-1</sup> h <sup>-1</sup>	64	47	44

<sup>1</sup>NDF = neutral detergent fibre, <sup>2</sup>ADF = acid detergent fibre, <sup>3</sup>ADL = acid detergent lignin, <sup>4</sup>INDF = indigestible NDF, <sup>5</sup> $k_d\text{DNDF}$  = degradation rate of digestible NDF.

Distributions of faeces particles in the different sieving fractions were significantly affected by cutting time (Table 2). Feeding early cut grass silage compared with late cut grass silage increased the proportion of particles retained in the lower fractions B and C and in the uppermost fraction O, but decreased the proportion of particles retained in the middle fractions S and D, leading to lower overall particle size values in terms of Mode\_PS, APS and GPS. The higher percentage of faecal particles longer than 2.36 mm from ewes fed early cut silage compared with those fed silages harvested at later maturities may be related to the higher DM intake (Tables 2 and 3). Similarly, Van Soest (1982) reported a linearly increasing faecal particle size, measured by wet sieving technique, at increasing intake by cattle fed forage. The higher proportions of small particles (< 0.212 mm) in faeces from ewes fed early cut grass silage may be related to less lignified tissue in the early cut silage that is easier to chew and masticate than more lignified tissue in later cut silages (Mertens, 1997). Cutting time of the silage also affected faecal DM content and DM loss during washing in nylon bags (Table 2). Number of LP > 10 mm was similar among treatments, averaging 26 LP 50 g<sup>-1</sup> faeces.

Table 2. Proportion (g kg<sup>-1</sup>) of faeces particles in the individual sieving fractions, overall mean particle size (mm), and faeces characteristics from ewes fed early cut (ECS), medium cut (MCS) and late cut (LCS) grass silages.

	ECS	MCS	LCS	SEM	P - value
<i>Sieving fraction</i>					
O, 2.36 mm	16.0 <sup>a</sup>	10.3 <sup>b</sup>	8.7 <sup>b</sup>	1.12	0.0008
M, 1.0 mm	13.4	13.3	9.2	2.01	ns
S, 0.5 mm	55.4 <sup>a</sup>	86.3 <sup>b</sup>	114.2 <sup>c</sup>	5.29	< 0.0001
D, 0.212 mm	204.1 <sup>a</sup>	314.0 <sup>b</sup>	410.2 <sup>c</sup>	26.20	0.0002
C, 0.106 mm	504.0 <sup>a</sup>	409.6 <sup>b</sup>	317.1 <sup>c</sup>	21.67	< 0.0001
B, Bottom bowl	203.0 <sup>a</sup>	166.5 <sup>b</sup>	140.6 <sup>c</sup>	10.32	0.0025
<i>Overall particle size</i>					
Mode_PS <sup>1</sup> , mm	0.107 <sup>a</sup>	0.207 <sup>b</sup>	0.256 <sup>c</sup>	0.012	< 0.0001
APS <sup>2</sup> , mm	0.263 <sup>a</sup>	0.286 <sup>ab</sup>	0.311 <sup>b</sup>	0.010	0.0203
GPS <sup>3</sup> , mm	0.181 <sup>a</sup>	0.203 <sup>a</sup>	0.228 <sup>b</sup>	0.007	0.0020
<i>Faeces characteristics</i>					
DM, g kg <sup>-1</sup>	269 <sup>a</sup>	307 <sup>b</sup>	346 <sup>c</sup>	13	0.0022
DM loss <sup>4</sup> , g kg <sup>-1</sup>	587 <sup>a</sup>	399 <sup>b</sup>	321 <sup>c</sup>	17	< 0.0001

<sup>a, b, c</sup>Means in the same row with different superscripts differ ( $P < 0.05$ ). ns = not significant,  $P > 0.10$ .

<sup>1</sup>Mode\_PS = most frequent particle size, <sup>2</sup>APS = arithmetic mean particle size, <sup>3</sup>GPS = geometric mean particle size, <sup>4</sup>DM loss = faecal dry matter loss during washing in nylon bags.

Table 3. Daily intake of ewes fed early cut (ECS), medium cut (MCS) and late cut (LCS) grass silages.

	ECS	MCS	LCS	SEM	P - value
BW, kg	93.7 <sup>a</sup>	90.4 <sup>ab</sup>	82.8 <sup>b</sup>	3.174	0.076
DM intake kg	2.5 <sup>a</sup>	2.1 <sup>a</sup>	1.5 <sup>b</sup>	0.150	0.001
DM intake, g kg <sup>-1</sup> BW	26 <sup>a</sup>	23 <sup>a</sup>	18 <sup>b</sup>	1.340	0.002
NDF <sup>1</sup> intake, kg	1.12	1.22	0.96	0.080	0.086
NDF intake, g kg <sup>-1</sup> BW	12	14	12	0.720	0.145
INDF <sup>2</sup> intake, g kg <sup>-1</sup> BW	0.9 <sup>a</sup>	2.4 <sup>b</sup>	3.3 <sup>c</sup>	0.132	0.001

<sup>a, b, c</sup>Means in the same row with different superscripts differ ( $P < 0.05$ ).

<sup>1</sup>NDF = neutral detergent fibre, <sup>2</sup>INDF = indigestible NDF.

## Conclusions

Stage of maturity of grass at harvest has a significant effect on faecal characteristics in pregnant ewes. Feeding early cut grass silage resulted in more small faeces particles, a lower overall mean particle size in faeces and a lower faecal DM content.

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# Organic milk production based entirely on grassland feeds

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## Abstract

A great part of the dairy cows' feed rations consists of grains and protein feeds which could be used for human consumption. Production of these feeds is less environmentally friendly than grassland production. How can high-producing dairy cows (Swedish Holstein) manage on perennial forages only? For three years 10 cows were fed organic grass-clover forages only (silage, pasture and small parts of hay), and were compared with 10 cows which were fed a 95% organic diet containing grass-clover forages, barley, peas and a commercial protein mixture. The concentrate proportion was, however, limited to 50% of the total mixed ration DM. Cows fed forages lost weight during the indoor periods and their plasma insulin levels decreased at the same time. However, cows fed forages only were healthy and fertile. They had an annual milk yield of around 6000 kg energy corrected milk (ECM) per cow compared to around 8700 kg ECM per cow when fed forages and concentrates. During the third year there were lower contents of protein and urea in milk from cows fed forages only. In conclusion, a high-producing dairy breed can manage well during extensive conditions.

Keywords: organic, milk production, grass-clover forages

## Introduction

Dairy production is based on forages and often high levels of grains and protein feeds. Concentrates could be used for human consumption and they are often expensive. Grassland production is the most environmental friendly crop production, and nitrogen-fixing legumes are needed in organic production. Several experiments have studied different levels of concentrates and shown decreased milk yields when feeding low levels of concentrates (e.g. Dhiman *et al.*, 1995; Sehestad *et al.*, 2003), but not many findings exist on how dairy cows cope on rations where the concentrates are omitted. Reducing feed intake levels in genetically superior dairy cows could have negative effects on health if it causes an increasing mobilization of body reserves and a more negative energy balance (Butler and Smith, 1989). The aim of this experiment was to study long-term effects on milk production, weight and body condition, health and energy balance when high-producing dairy cows were fed perennial grass-clover forages only.

## Materials and methods

The experiment was conducted during three years at Tingvall Organic Research Farm, which was owned by the Rural Economy and Agricultural Societies, Sweden. The herd consisted of Swedish Holstein Cows with lactation number 1-7 and calving date between October and December. The cows were milked twice a day and were housed in a loose housing system. Twenty cows were paired according to expected calving date and lactation number, and then randomly allocated to the two treatments. The treatments were F: Forages only, consisting of grass-clover silage, hay and pasture, and C: Control, a total mixed ration of grass-clover silage, hay, barley, peas and commercial protein concentrates. During the third year, rapeseed

cake also was included in the ration. The feeding strategy for C followed the standards for organic production (KRAV, 1997; KRAV 2000) with a maximum of 50% concentrates. Samples from the feeds were taken continuously and analysed for chemical composition (Analycen AB, Lidköping, Sweden). All compositions are shown in Johansson *et al.* (2002). The silage in year three contained dry matter (DM): 26 g kg<sup>-1</sup>, metabolizable energy: 10.3 MJ kg<sup>-1</sup> DM, crude protein: 129 g kg<sup>-1</sup> DM and NDF (neutral detergent fibre): 505 g kg<sup>-1</sup> DM. Milk yield was recorded once a month and also 6, 12, 18, 21 and 22 weeks after calving. Energy corrected milk was calculated as defined by Sjaunja *et al.* (1990). Milk composition was analysed by Steins Laboratory AB (Jönköping, Sweden). Body weight was recorded once a month, and body condition of the cows was scored by one trained scorer, using a visual appraisal method (Edmonson *et al.*, 1989). During the third year, blood samples were collected from the cows every second week from calving to six months after calving. The blood plasma was analysed for content of insulin with a radioimmunoassay technique (Pharmacia Diagnostics AB, Uppsala, Sweden). Because effect on energy balance (insulin) was studied during year three, all statistical analyses shown here are based on results from the third year. Statistical analyses of variance were made using the Mixed Model procedure of SAS (1999). The statistical model included effect of treatment. Results with a *P* value less than 0.05 were regarded as statistically significant.

## Results and discussion

The annual milk yield of cows fed forages only averaged 5,700, 5,800 and 6,350 kg cow<sup>-1</sup> compared to 8,550, 8,000 and 9,500 kg for control cows in years 1, 2 and 3, respectively. Also, daily kg milk and ECM yields cow<sup>-1</sup> were lower for F-cows during both early and later lactation (Table 1). In agreement with Sehestad *et al.* (2003) the milk-protein content was reduced for F-cows compared with C-cows; however, the effect was greater in later lactation. Also, content of milk urea was lower for F-cows than for C-cows. Low urea levels might affect fertility negatively, but this was not seen in this study (Johansson *et al.*, 2002).

Table 1. Milk yield, milk composition and plasma level of insulin during early (0-90 days) and later (91-270 days) lactation for cows fed forages only (F) or for control (C) cows. Least square means and standard error of the difference between means (SED).

Lactation stage (days)	0-90			91-270 (90-180 for insulin)		
	F	C	SED	F	C	SED
Milk (kg)	23.8	35.2***	2.1	19.0	28.1***	1.9
ECM <sup>1</sup> (kg)	22.3	34.6***	2.4	18.0	28.8***	1.8
Fat (g kg <sup>-1</sup> )	37.5	39.8	2.3	38.4	42.6	3.0
Fat (kg)	0.90	1.39***	0.11	0.74	1.19***	0.08
Protein (g kg <sup>-1</sup> )	29.4	32.3*	1.1	29.9	34.2***	1.0
Protein (kg)	0.70	1.13***	0.08	0.58	0.96***	0.07
Urea (mM)	2.49	3.36*	0.31	2.86	3.59*	0.27
Insulin (μUml <sup>-1</sup> )	8.65	13.21***	1.14	10.48	14.27***	1.12

\*, \*\*\*, Significant differences between two treatment means within the same row and lactation stage (\**P* < 0.05, \*\*\**P* < 0.001).

Level of insulin in plasma was lower for F-cows than for C-cows during all lactation stages but the difference was larger during the first three months after calving than for later lactation (Table 1). When cows are producing high amounts of milk in early lactation the risk of a negative energy balance is high. Low levels of insulin stimulate feed intake and facilitate mobilization of body fat and protein. Body weight and body condition score followed the insulin level well in this study. The weight and score was lower for F-cows during wintertime than for C-cows, but increased when cows were let out on pasture (Figure 1). In spite of large

losses in weight and body condition, the health was better for F-cows than for C-cows (Johansson *et al.*, 2002). A lower treatment frequency in herds with low production levels was also found earlier (e.g. Hardeng *et al.*, 2001; Sehestad *et al.*, 2003).

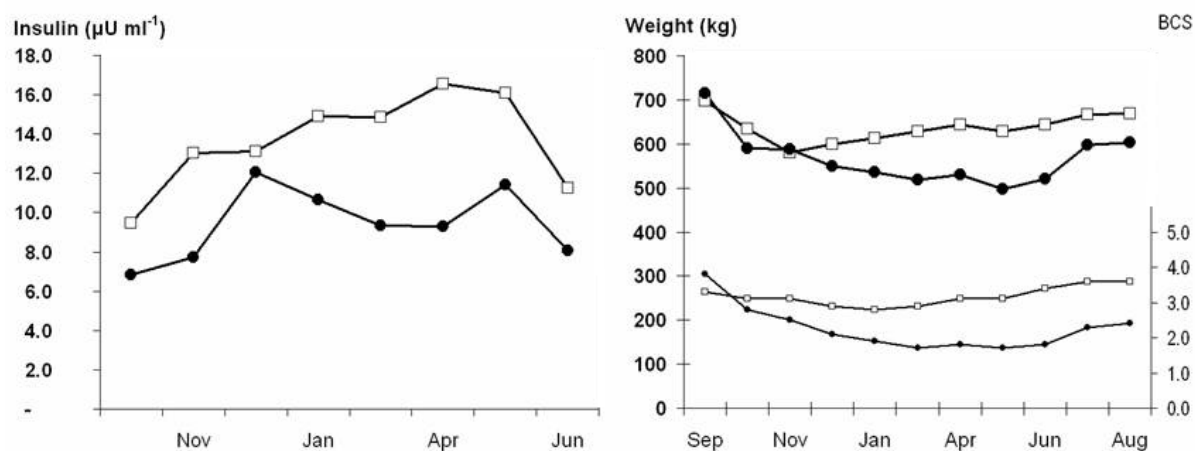


Figure 1. Level of insulin in blood plasma, weight and body condition score (BCS) during year three, for cows fed forages only (●) and control cows (□).

## Conclusions

High-producing dairy cows adapt well to extensive conditions. Cows fed only forages lost weight and had low levels of insulin in blood, but were healthy and fertile. They also produced more milk than expected. However, due to the low body condition score, forages should be supplemented with some concentrates, especially in high-yielding cows in early lactation.

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## Reverting to grazing: farmers' conceptions

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### Abstract

The Niort plain (Poitou-Charentes, France) presents several environmental challenges due to the presence of: (i) the protected bird species, Little Bustard (*Tetrax tetrax*), which needs different types of grassland for its optimal development (nesting and growth), and (ii) droughts which reduce the water available for maize irrigation. This shortage of irrigation water for maize means that cattle tend to be grazed rather than being fed silage. Both these factors make for a renewed interest in grassland. In this context, socio-anthropological interviews were carried out with eleven farmers to ascertain their views about grazing for dairy cows, and whether or not they practise it. Results of interviews are presented in terms of farmers' general ideas on grazing; its implications for product (butter) labelling; their conceptions in terms of labour requirements and herd management; and the security and simplicity of diets (erratic milk production associated with transition from maize silage to grazing in spring, and implications of effects on rumen flora). Labour specialization, linked to farm enlargement, is seen to lead to greater complexity, and grazing management is seen by farmers as complex, compared with the stability and simplicity of a maize diet.

Keywords: grassland, grazing, conception of grazing by farmers, obstacles to the grazing reintroduction

### Introduction

In the Niort plain (Poitou-Charentes, France), several environmental factors (e.g. the presence of a protected bird, the Little Bustard (*Tetrax tetrax*) and drought, which reduces the scope for maize irrigation), are leading researchers and farmers to try to replace the maize crop with grassland, and a maize silage diet for dairy cows with grazing. In a research programme we have studied, using socio-anthropological methods, what farmers think of grazing and whether their cows graze or not. In this way we can learn about local conceptions of grazing, although grazing has quite disappeared in the region since 20 years.

### Materials and methods

The approach is linked to Weber's comprehensive sociology (Weber, 1971). Putnam (1984) states that a social group creates a system of thinking which is the result of breaking down reality due to its own experience. The linguist Prieto (1975) shows that a social group creates categories within the reality, gives a word for each category and puts objects into them. Through individual interviews performed in a locality (2-3 villages), and an analysis of these discussions, we can find out how the farmers of this locality conceive dairy cow grazing (Darré *et al.*, 2004).

Here, we worked on two localities of three villages. As the results were similar for both, we will not distinguish between them. Eleven individual interviews were performed with different farmers (farm area, age, grazing or not). Interviewed farmers are encouraged to respond to three questions: Can you talk about (i) the way you and your neighbours produce milk, (ii) the recent changes in the way you and your neighbours feed animals, (iii) advantages and difficulties of dairy cow grazing? Farmers have the choice of the content of the discourse.

The analysis of the discussions consists in identifying the different issues within each interview. For each issue, we study how farmers are talking about things or actions, by studying the words associated and opposed, and how things or actions are qualified. A summary of each interview is made. A global summary lists the most important points of the debate.

## **Results**

### *General ideas on grazing*

Farmers think that grazing is good for animals. They like to see cows grazing in pastures, and think cows feel better there than indoors. Out of doors, cows have fewer problems with lameness and are more contented.

However grazing is associated with the past, and their forefathers, so reverting to grazing is regarded as a retrograde step.

Grazing is considered to be economically profitable by the farmers who practise it; the others ask whether the benefits are significant and would like to have local studies on this topic.

### *Importance of the label of origin of the product (Echiré butter)*

The position of farmers is paradoxical: they think that grazing would add to the value of the labelled product: it provides a good image for the consumer. But they consider that, as few farmers graze, grazing should not be specified on the label. Moreover, butter prices are higher from July to Christmas, whereas the grazing period is during spring.

### *Farm structures and farm functioning*

For farmers, grazing is possible only if the cowshed is close to the pastures. They do not like to drive their cattle along roads, especially close to towns. Big herds are difficult to lead, and cattle are liable to escape. Moreover, the management of such herds by several associates raises new problems: are all the associates able to manage the grazing when their turn comes? Farmers think that the labour requirement is much the same for grazing and for maize silage feeding, but that the tasks are different. Grazing requires more flexibility in herd management and in work distribution. Farmers who practise grazing enjoy being out of doors, keeping an eye on the grass and the animals.

### *Security and simplicity of diets*

A basic diet of maize silage is regarded as reliable and results in regular milk production. It is simple and easy to manage.

By comparison, grazing cows have erratic milk production and management is not so easy because of the variations in grass quality. During the beginning of the season (March), grass grows quickly but 'by the 15th of May there is no more grass'. The grazing period is short. Farmers are worried about the change of diet. They say that 'the transition from maize silage to grazing need to be gradual: the rumen flora has to get used to it'. Dietary changes lead to a modification of the entire organism: perturbations provoke a decrease of milk production, and may be the cause of other problems, e.g. with fecundity. For these reasons, farmers don't want to close the maize silo during grazing period.

Some farmers who are getting a high production per cow do not want production to fall, and are afraid to try grazing.

Grazing needs particular techniques and anticipation: determination of the paddock size and of the rhythm of the grazing rotation. An indicator of the need to change the paddock is the extent to which animals leave maize silage in the rack. Learning grazing management is difficult for farmers. They say that it is not a practice they learned at school or in books, so if their father did not practise it, they have to learn from neighbours.



## Discussion

In this section, we put farmers' conceptions into perspective with advisers' and researchers' opinions. For the dairy industry, grazing provides a good image for the product. But the price of butter is higher towards the winter festive season than during the grazing season. Furthermore, butter from grazing cows is yellow, but for the label's reputation it should be white, so the industry doesn't want to promote grazing.

A huge disadvantage for grazing is that farmers want all the pastures to be near the cowshed. With this constraint, to promote grazing would need spatial reorganization of the fields.

The fear of dietary changes seems to be exaggerated by farmers. Delaby and Peyraud (2002) showed there are no effects on milk production when ration transitions are well managed. But to graze, farmers would have to accept irregular production. Our economic study of farms which have just introduced grazing, or are thinking of introducing it, shows that the changeover to grazing benefits the farm.

An adviser remarks that when the diet consists of both grazing and silage it is not balanced, and knowing how much silage to give is not easy. This twin-component diet seems to provide security, but is perhaps very expensive.

Farmers lack knowledge about grazing management: new research is needed to provide this knowledge.

## Conclusions

A challenge is to show that grazing is not just a practice from the past, but one worth reinventing. However, we have seen that its promotion needs to greatly modify farm structures, farmers' thinking and even, through knowledge, tasks of advisers and researchers.

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## Previous nutrition and entry time to pasture affect weight gain and grazing behaviour in steers

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### Abstract

Sixteen Holstein steers ( $139 \pm 12$  kg body weight (BW)) were used in a randomized block, 2 x 2 factorial, 64-d experiment, to assess the effect of previous nutrition (PN) and entry time (ET) to a winter *Avena sativa* pasture on daily weight gain (DWG) and grazing (G) and ruminating (R) time. Steers were initially fed *Lolium perenne* and *Medicago sativa* hay, for 21 d, to gain  $8 \pm 43$  (L; N = 8) or  $345 \pm 92$  (H; N = 8) g BW d<sup>-1</sup>, and then allowed to graze individual paddocks of winter *Avena sativa* pasture (one-day strip grazing) for 29 d. Entry time to the paddocks was at 8:30 (M; N = 8) or 14:30 (A; N = 8) h. Steers were weighed on days 28 and 64 to estimate DWG. Grazing time and R were estimated over 24 h using IGER behaviour recorders from days 51 to 60. Daily weight gain was affected by PN (564 (L) vs. 675 (H) g d<sup>-1</sup>;  $P = 0.074$ ) and ET (573 (M) vs. 668 (A) g d<sup>-1</sup>;  $P = 0.077$ ). Entry time altered G (425 (M) vs. 514 (A) min d<sup>-1</sup>;  $P = 0.012$ ) and PN affected R (438 (H) vs. 520 (L) min d<sup>-1</sup>;  $P = 0.02$ ).

Keywords: entry time, level of nutrition, grazing behaviour, weight gain, steers, winter oats

### Introduction

In the evening, the grass has a comparatively higher nutritional value for ruminants, with higher contents of dry matter (DM) and non-structural carbohydrates (NSCH) and lower contents of neutral detergent fibre (NDF) and crude protein (CP) (Delagarde *et al.*, 2000). Thus, in one-day strip grazing systems, an improvement in performance would be expected if animals were given access to a new strip of pasture in the afternoon rather than in the morning (Gregorini *et al.*, 2006). Oat (*Avena sativa*), widely used as winter grass in rotational systems in Argentina, has been reported to have high CP / NSCH ratios (Martínez *et al.*, 2005). This may result in high N losses from the rumen and high portal NH<sub>4</sub><sup>+</sup> flow that exceed liver NH<sub>4</sub><sup>+</sup> uptake and increase peripheral blood NH<sub>4</sub><sup>+</sup>. In sheep, small additions of non-protein N to the morning meal resulted in high plasma NH<sub>4</sub><sup>+</sup> concentrations in peripheral blood, and marked reductions in N retention (Milano and Recavarren, 2007). This may be overcome by a high plane of nutrition, which increases liver mass (Burrin, 1990) and liver capacity for NH<sub>4</sub><sup>+</sup> uptake. The objective of this experiment was to assess the effect of previous nutrition (PN) and entry time (ET) to an oat pasture on daily weight gain (DWG), grazing (G) and ruminating (R) time and plasma NH<sub>4</sub><sup>+</sup> in steers during late autumn-early winter.

### Materials and methods

*Animals, diets and design.* Sixteen Holstein steers were used in a 64-d experiment (8/05/07 to 10/07/07) with 2 periods, and a 10-d interval. Initially, the steers were weighed (W0), following a 48-h fasting period, and allocated to feeding groups L (N = 8;  $142 \pm 12$  kg body weight (BW)) or H (N = 8;  $135 \pm 12$  kg BW). From d 1 to d 21 (period 1), steers in group L were offered 1.8 kg DM d<sup>-1</sup> of *Lolium perenne* hay (92 g CP kg DM<sup>-1</sup>; 634 g NDF kg DM<sup>-1</sup>) and 1.8 kg DM d<sup>-1</sup> of *Medicago sativa* hay (179 g CP kg DM<sup>-1</sup>; 423 g NDF kg DM<sup>-1</sup>) per

animal, to maintain BW. Steers in group H were offered 2.6 kg DM d<sup>-1</sup> of *Lolium perenne* hay, 2.6 kg DM d<sup>-1</sup> of *Medicago sativa* hay and 0.4 kg DM d<sup>-1</sup> of cracked *Zea mays* per animal, to gain ca 400 g BW d<sup>-1</sup>. During the 10-d interval (d 22 to d 31), the steers grazed, firstly, a common paddock of winter *Avena sativa* pasture (one-day strip grazing; forage allowance, 56.5 g DM kg BW<sup>-1</sup> d<sup>-1</sup>; ET, 12:00 h) for 4 d, were then fasted for 2 d, weighed (W1), and finally allowed to recover from fasting on a *Lolium perenne* pasture (continuous grazing) for 4 d. The steers were arranged in four blocks, each block having two pairs of steers of similar BW according to W1, with each pair within each block coming from group L and H, respectively. From d 32 to d 61 (period 2), the steers grazed individual paddocks of winter *Avena sativa* pasture (7181 kg DM ha<sup>-1</sup>; 138 g DM kg<sup>-1</sup>; 150 g CP kg DM<sup>-1</sup>; 497 g NDF kg DM<sup>-1</sup>; 295 g ADF kg DM<sup>-1</sup>; 165 g NSCH kg DM<sup>-1</sup>; 922 g OM kg DM<sup>-1</sup>; sampled at ca. 12:00 h), under a one-day strip grazing system (forage allowance, 56.5 g DM kg BW<sup>-1</sup> d<sup>-1</sup>). ET to the paddocks for each member of the pair was either at 8:30 (M; N = 8) or 14:30 (A; N = 8) h. Water was always offered *ad libitum* except during period 2, when steers had no access to water because expected water intake with the forage (ca. 170 g kg BW<sup>-1</sup> d<sup>-1</sup>), based on water content of the forage (862 g kg fresh forage<sup>-1</sup>) and estimated forage DM intake (30 g DM kg BW<sup>-1</sup> d<sup>-1</sup>), exceeded water requirements (90 g kg BW<sup>-1</sup> d<sup>-1</sup>).

*Daily weight gain.* Steers were weighed on d 1 (W0), d 28 (W1) and d 64 (W2), after a 48-h fast, to estimate DWG over each experimental period.

*Herbage mass and dry matter intake (DMI).* Initial herbage mass was determined on d 21 and d 52, by clipping to ground level eight squares of 0.075 m<sup>2</sup> in ungrazed paddocks. On d 64 and d 66, samples of residual herbage mass were obtained from all paddocks grazed on d 60 and d 61 by clipping to ground level eight squares of 0.075 m<sup>2</sup> per paddock. Forage samples were oven dried at 95-100 °C and individual DMI was calculated as the difference between average initial herbage mass on d 52 and residual herbage mass.

*Blood samples.* On d 36, blood samples from jugular vein were collected from all steers into heparinised syringes 120 min after entering the pasture (i.e., 10:30 h for group M and 16:30 for group A) and immediately analysed for plasma NH<sub>4</sub><sup>+</sup>.

*Grazing behaviour.* Grazing and ruminating behaviour were recorded automatically over 24 h using four IGER Grazing Behaviour Recorders (Ultra Sound Advice, UK), on alternate days, from d 51 to d 60. The recorders were fitted to the animals at ca. 8:00 h, with recordings starting, in all cases, at ca. 8:10 h and ending 24 h later. Each steer was recorded once during the experiment. Recordings were analysed using the GRAZE<sup>TM</sup> Software (Ultra Sound Advice, UK) to determine total time spent grazing (G; included biting and non-biting jaw movements and intrameal intervals < 5 min) and ruminating (R; included ruminating jaw movements and intervals between boluses).

*Statistics.* Variates were analysed by ANOVA (SAS 9.1, 2007), as a 2 x 2 factorial, with PN (L or H) and ET (M or A) as main effects, and W1 as covariate.

## Results and discussion

Results are shown in Table 1. In the first period, DWG for feeding groups L and H was 8 ± 43 and 345 ± 92 g BW d<sup>-1</sup> ( $P < 0.001$ ; Student *t* Test), respectively. Feeding steers at contrasting levels of nutrition (1.0 vs. 1.5 maintenance energy intake) for 21 d previous to grazing aimed at increasing liver mass and capacity for portal NH<sub>4</sub><sup>+</sup> removal, to prevent any excess NH<sub>4</sub><sup>+</sup> absorbed from the rumen at times when CP / NSCH ratio is high from reaching peripheral blood. However, in all treatments, plasma NH<sub>4</sub><sup>+</sup> was below that associated with low N retention (200 µM; Milano and Recavarren, 2007), suggesting CP and / or CP / NSCH ratio in the grazed forage were never high enough to increase NH<sub>4</sub><sup>+</sup> absorption beyond liver capacity for NH<sub>4</sub><sup>+</sup> uptake. Nevertheless, DWG was 111 g d<sup>-1</sup> higher in H steers ( $P = 0.074$ ), although this was unrelated to DMI or G. Only R was 72 min shorter in H ( $P = 0.036$ ), but its relation

with DWG is not clear. In the second period, DWG was 95 g d<sup>-1</sup> higher for steers with ET at 14:30 h ( $P = 0.077$ ). Similar results (+ 150 g d<sup>-1</sup>) were reported by Gregorini *et al.* (2006) with heifers grazing *Lolium multiflorum* pasture in winter, with ET at 7:00 and 15:00 h. However, rather than the 60 min reduction in daylight G seen in that experiment, our recordings showed no change in G during daylight. Instead, an increase in 24-h G (55 min,  $P = 0.042$ ), due to longer G during night hours (112 min;  $P < 0.001$ ), was observed for animals with ET at 14:30 h. Dry matter intake, estimated by a coarse methodology, was not altered by ET or PN. However, pending results from the *n*-alkane technique, which was also used to calculate DMI in this experiment, could show otherwise. Alternatively, changes in DWG might be related to the coordination of changes in forage composition (Delagarde *et al.*, 2000) with the pattern of grazing bouts during the day.

Table 1. Body weight (W1), daily weight gain (DWG), dry matter intake (DMI), plasma NH<sub>4</sub><sup>+</sup> and grazing (G) and ruminating (R) time in steers grazing winter *Avena sativa* pasture at two different entry times (ET; M, 8:30 and A, 14:30 h) and with two levels of nutrition previous to grazing (PN; L, + 8 g BW/d; H, + 345 g BW/d).

	PN		ET		RMSE	<i>P</i>		
	L	H	M	A		PN	ET	PNxET
W1(kg)	143	144	143	143	12.8	0.932	0.947	0.977
DWG <sub>d32-61</sub> (g d <sup>-1</sup> )	564	675	573	668 <sup>a</sup>	143.6	0.074	0.077	0.339
DMI (g d <sup>-1</sup> )	4,861	4,971 <sup>a</sup>	4,880	4,967 <sup>a</sup>	734.9	0.764	0.879	0.419
G <sub>24</sub> (min) <sup>bc</sup>	471	438	425	480	40.3	0.257	0.042	0.183
G <sub>8:30-18:30</sub> (min) <sup>d</sup>	308	301	329	272	32.7	0.659	0.141	0.201
G <sub>18:30-8:30</sub> (min) <sup>d</sup>	164	138	96	208	24.1	0.272	< 0.001	0.853
R <sub>24</sub> (min) <sup>bc</sup>	505	433	476	439	43.3	0.036	0.340	0.106
R <sub>8:30-18:30</sub> (min) <sup>d</sup>	125	92	103	104	25.6	0.080	0.793	0.164
R <sub>18:30-8:30</sub> (min) <sup>d</sup>	381	341	372	335	50.2	0.090	0.161	0.225
NH <sub>4</sub> <sup>+</sup> (μM)	44	38	41	43	8.6	0.346	0.725	0.643

<sup>a</sup>N = 7. <sup>b</sup>G<sub>24</sub> and R<sub>24</sub>. 24-h recordings of G and R. <sup>c</sup>Eleven complete 24-h recordings obtained (N<sub>L</sub> = 4, N<sub>H</sub> = 7, N<sub>M</sub> = 6, N<sub>A</sub> = 5). <sup>d</sup>G and R over indicated interval.

## Conclusions

Daily weight gain increased in young Holstein steers when ET to a winter *Avena sativa* pasture was switched from 8:30 to 14:30 h ( $P = 0.077$ ), or when they were fed at a higher PN (1.5 vs. 1.0 maintenance) for 21d previous to grazing ( $P = 0.074$ ). Plasma NH<sub>4</sub><sup>+</sup> was unaltered by treatments. Grazing time was longer for steers with ET at 14:30 h, due to longer G at night ( $P < 0.001$ ).

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# Performance of pregnant and lactating ewes fed grass silages differing in maturity

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## Abstract

The effect of grass silage (timothy, meadow fescue, perennial ryegrass) maturity on performance of pregnant and lactating ewes and on live weight (LW) gains of their lambs during the suckling period, was studied. Eighteen Swedish Finull x Dorset ewes were fed early (E), medium (M) or late (L) cut grass silage containing 11.7, 10.8 and 9.3 MJ metabolizable energy and 472, 620 and 665 g NDF kg<sup>-1</sup> DM, respectively. The ewes were fed 0.8 kg concentrate during late pregnancy and lactation. Silage DM intake was 23.4, 18.2 and 16.6 g kg<sup>-1</sup> LW during late pregnancy and 37.3, 30.1 and 27.8 g kg<sup>-1</sup> LW during lactation of E, M and L silage, respectively ( $P < 0.01$ ). The LW and body condition of the ewes fed E, M and L silage were 102, 99, 90 kg and 3.1, 2.8, 2.5 ( $P = 0.01$ ), respectively, during late pregnancy, and 96, 87, 79 kg and 2.9, 2.3, 1.9, respectively, during lactation ( $P < 0.01$ ). Daily LW gain of the lambs was 421, 381 and 353 g ( $P = 0.002$ ) born from ewes fed E, M and L silages, respectively. Feeding grass silage at early maturities improves performance of pregnant and lactating ewes, which results in increased LW gain of their sucking lambs.

## Introduction

Maturity stage at harvest is one of the most important factors affecting forage quality (Nelson and Moser, 1994). Increased fibre concentration but decreased fibre digestibility with delayed maturity of forages at harvest increase rumen fill, which limits intake by ruminants with high energy demands (Mertens, 1994; Allen, 1996). Animals that have energy intakes that are too low in comparison to their requirements use body reserves for energy release. Large energy deficiencies of pregnant and lactating ewes affect not only their own performance but also the performances of their lambs. The objective of this experiment was to determine the effect of grass silage maturity on the performance of pregnant and lactating ewes and on live weight (LW) gains of their lambs during the suckling period.

## Materials and methods

Eighteen Swedish Finull x Dorset ewes were fed early (E), medium (M) or late (L) cut grass silage, containing timothy (*Phleum pratense* L.), meadow fescue (*Festuca pratensis* L.) and perennial ryegrass (*Lolium perenne* L.), during pregnancy and lactation at Götala Research Station, Skara, Sweden. In 2006, the E, M and L grass was harvested on the 2nd, 12th and 21st of June, respectively, during the spring growth cycle and ensiled as long grass in round bales. Chemical composition of the silages was analysed by conventional methods and metabolizable energy was calculated from *in vitro* rumen organic matter disappearance (Table 1). The ewes were fed 0.8 kg concentrate in late pregnancy and lactation and the lambs were fed concentrate *ad libitum* in addition to the forage. Feed intake was registered daily. The ewes and their two lambs each were weighed once a week and the ewes were scored for body condition. The ewes were randomly assigned to the three silage treatments with six ewes per treatment. The average live weight (LW) of the ewes at the start of the experiment, which occurred  $44 \pm 1$  d. pre-partum, was 82.0 (SD 5.9), 82.3 (SD 4.9) and 83.5 (SD 4.2) kg for early, medium and late cut silage, respectively. The corresponding body condition scores were



3.3 (SD 0.3), 3.4 (SD 0.4) and 3.4 (SD 0.5). Data from the twelve lambs per treatment were analysed as an average across gender. Data on ewes during early and late pregnancy and lactation were analysed separately in a completely randomized design using the general linear model procedure of SAS (ver. 9.1, 2001). Data on lambs were analysed using the mixed procedure of SAS (ver. 9.1, 2001), where treatment and gender were fixed effects and twin lambs nested within treatment was a random effect. Least square means were used to account for the uneven distribution of gender of the lambs within treatment.

Table 1. Chemical composition, kg<sup>-1</sup> DM, unless stated otherwise, of early, medium and late cut silages fed to ewes<sup>1</sup>.

	Early cut silage		Medium cut silage		Late cut silage	
	Mean	SD	Mean	SD	Mean	SD
DM, g kg <sup>-1</sup>	484	39	586	46	524	67
Crude protein, g	177	12	119	20	92	14
NDF, g	472	20	620	14	665	21
Sugar, g	202	0	139	26	117	9
Metabolizable energy, MJ	11.7	0.3	10.8	0.5	9.3	0.3
<i>In vitro</i> rumen OMD, g kg <sup>-1</sup>	908	18	834	35	738	22
Lactic acid, g	10.3	13.2	4.6	5.3	18.4	4.8
Acetic acid, g	1.6	1.8	1.6	1.2	5.7	0.4
Butyric acid, g	< 0.19	0.005	< 0.16	0.009	< 0.21	0.012
Ethanol, g	32	1.9	17	0.7	20	4.0
Ammonium-N, g kg <sup>-1</sup> total N	56	1.4	24	12	87	14
pH	5.4	0.2	5.5	0.2	5.1	0.2

<sup>1</sup>n = 5 for DM, crude protein, NDF, metabolizable energy and *in vitro* rumen organic matter disappearance (OMD); n = 2 for sugar and fermentation quality.

## Results and discussion

During early pregnancy, when no concentrate was fed, the dry matter (DM) intake of early cut silage was 16 and 49% higher than that of medium and late cut silages, respectively (Table 2). Also, the DM intake of early cut silage was, on average, 34% and 29% higher than that of medium and late cut silages during late pregnancy and lactation, respectively (Table 3). There was no difference in NDF intake among the silages, except for a lower NDF intake of late cut silage than of early and medium cut silages in early pregnancy (Tables 2 and 3).

Table 2. Silage intake, kg<sup>-1</sup> live weight (LW), LW and body condition score (BCS) of ewes during early pregnancy (n = 6).

	Early pregnancy <sup>1</sup>			SEM	P - value
	E <sup>2</sup>	M <sup>2</sup>	L <sup>2</sup>		
DM, g	27.6 <sup>a</sup>	23.7 <sup>b</sup>	18.5 <sup>c</sup>	1.09	0.0001
NDF, g	13.8 <sup>a</sup>	14.4 <sup>a</sup>	11.8 <sup>b</sup>	0.61	0.02
Crude protein, g	4.2 <sup>a</sup>	2.6 <sup>b</sup>	1.9 <sup>c</sup>	0.15	< 0.0001
ME <sup>3</sup> , MJ	0.31 <sup>a</sup>	0.25 <sup>b</sup>	0.18 <sup>c</sup>	0.01	< 0.0001
LW <sup>4</sup> , kg	91	88	85	2.7	ns
BCS	3.4	3.1	3.0	0.14	ns

<sup>1</sup>25 d. long, <sup>2</sup>E, M and L are early, medium and late cut silages, respectively.

<sup>3</sup>ME = metabolizable energy in Megajoule calculated from *in vitro* organic matter disappearance.

<sup>4</sup>LW includes the weights of two fetuses.

<sup>a-c</sup>Means with different superscripts within a row differ significantly at  $P < 0.05$ . ns = not significant,  $P > 0.10$ .

As DM intake as well as concentrations of crude protein and ME decreased, intakes of these components decreased with later harvest date of the grass. The decreasing ME intake resulted in decreasing BCS of the ewes in late pregnancy and lactation with later harvest time of the grass (Table 3). Also, the LW of the ewes were lower for medium and late cut silages than for

early cut silage during lactation. Ewes fed the late cut silage lost too much body condition during lactation. Results from a field study on well managed ewes show BCS of 3.0 at parturition with a decrease to 2.5 during lactation (Arnesson and Eggertsen, 2006).

Table 3. Silage intake, kg<sup>-1</sup> live weight (LW), LW and body condition score (BCS) of ewes during late pregnancy and lactation (n = 6).

	Late pregnancy <sup>1</sup>			SEM	P-value	Lactation <sup>2</sup>			SEM	P-value
	E <sup>3</sup>	M <sup>3</sup>	L <sup>3</sup>			E	M	L		
DM, g	23.4 <sup>a</sup>	18.2 <sup>b</sup>	16.6 <sup>b</sup>	0.14	0.009	37.3 <sup>a</sup>	30.1 <sup>b</sup>	27.8 <sup>b</sup>	1.14	< 0.0001
NDF, g	10.9	10.8	11.2	0.73	ns	17.5	17.8	17.7	0.65	ns
Crude protein, g	4.2 <sup>a</sup>	2.7 <sup>b</sup>	1.4 <sup>c</sup>	0.21	< 0.0001	7.0 <sup>a</sup>	3.4 <sup>b</sup>	3.2 <sup>b</sup>	0.20	< 0.0001
ME <sup>4</sup> , MJ	0.28 <sup>a</sup>	0.20 <sup>b</sup>	0.15 <sup>b</sup>	0.01	0.0003	0.44 <sup>a</sup>	0.33 <sup>b</sup>	0.27 <sup>c</sup>	0.01	< 0.0001
LW <sup>5</sup> , kg	102	99	90	3.18	0.06	96 <sup>a</sup>	87 <sup>b</sup>	79 <sup>b</sup>	3.10	0.004
BCS	3.1 <sup>a</sup>	2.8 <sup>b</sup>	2.5 <sup>b</sup>	0.12	0.01	2.9 <sup>a</sup>	2.3 <sup>b</sup>	1.9 <sup>c</sup>	0.11	0.0001

<sup>1</sup>19 ± 1 d pre partum, <sup>2</sup>48 ± 1.5 d. long, <sup>3</sup>E, M and L are early, medium and late cut silages, respectively.

<sup>4</sup>ME = metabolizable energy in Megajoule calculated from *in vitro* organic matter disappearance.

<sup>5</sup>LW during pregnancy includes the weights of two foetuses.

<sup>a-c</sup>Means with different superscripts within a row differ significantly at  $P < 0.05$ . ns = not significant,  $P > 0.10$ .

Lambs born from ewes fed medium and late cut silages had lower LW gains during their suckling period than lambs born from ewes fed early cut silage (Table 4). The differences in LW gain resulted in lower LW at weaning for lambs from ewes fed late cut silage compared to lambs from ewes fed early cut silage. Lambs from ewes fed early, medium or late cut silage consumed 5.8, 6.8 and 8.2 kg concentrate, respectively, during the whole suckling period.

Table 4. Live weights (LW) at birth and at weaning and daily live weight gains of lambs during the suckling period (48 ± 1.5 d. long); n = 12.

	E <sup>1</sup>	M <sup>1</sup>	L <sup>1</sup>	SEM	P-value
LW at birth, kg	5.3	4.9	4.9	0.21	ns
LW at weaning, kg	25.9 <sup>a</sup>	23.1 <sup>a,b</sup>	21.8 <sup>b</sup>	1.05	0.03
LW gain, g d <sup>-1</sup>	421 <sup>a</sup>	381 <sup>b</sup>	353 <sup>b</sup>	11.7	0.002

<sup>1</sup>E, M and L are early, medium and late cut silages, respectively.

<sup>a,b</sup>Means with different superscripts within a row differ significantly at  $P < 0.05$ . ns = not significant,  $P > 0.10$ .

## Conclusion

Feeding grass silage harvested at an early maturity stage improves intake and performance of pregnant and lactating ewes, which results in increased LW gain of their sucking lambs.

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# Influence of silage structure on feeding behaviour and abnormal behaviours in dairy heifers

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## Abstract

The combined effects of maturity and particle size (PS) and the single effect of PS in grass silage on feeding behaviour and abnormal behaviours in dairy heifers were investigated. The study included two trials, where early cut (11.6 MJ, 510 g NDF kg<sup>-1</sup> DM) grass silage with a PS of 231 ( $\pm$  147) mm was compared to a late cut (10.0 MJ, 643 g NDF kg<sup>-1</sup> DM) silage with a PS of 373 ( $\pm$  245) mm in trial 1, whereas grass silages (534 and 540 g NDF kg<sup>-1</sup> DM) with two PS, 26 ( $\pm$  8.3) vs. 254 ( $\pm$  40) mm, were compared in trial 2. Both trials were conducted as a change-over design. Heifers spent less time eating but more time ruminating when fed the early cut and short silages compared to the late cut and long silages. However, heifers spent more time being idle when fed the early cut and short silages. These differences in eating behaviours resulted in higher probabilities for abnormal behaviours, such as licking the fixture, when heifers were fed the early cut and the short silages. The percentages of observations of abnormal behaviours were less than 2% for both treatments and did not affect the performance of the heifers.

Keywords: forage maturity, silage particle size, feeding, behaviour

## Introduction

Modern dairy production has come a long way when it comes to accommodating the nutritional and physiological needs of the animals but, unfortunately, the behavioural and psychological needs of cows and heifers are often left unfulfilled (Redbo and Nordblad, 1997). Chewing is necessary for a balanced rumen fermentation of feed particles and keeps the cattle active, leaving less time for developing abnormal behaviours. The most important factors influencing feed intake and chewing behaviour in cattle are particle size and fibre digestibility of forages and the content of forage fibre in the ration (Beauchemin *et al.*, 1994; Rinne *et al.*, 2002). The aim of this study was to investigate the combined effects of maturity and particle size (PS) and the single effect of PS in grass silage on feeding behaviour and abnormal behaviours in dairy heifers.

## Materials and methods

The study included two trials. Trial 1 was performed on a private dairy farm near Skara. Fifty-two Swedish Friesian (SF) heifers were divided into two groups: a younger group with 25 heifers aged 3-7 months, weighing 134 ( $\pm$  36) kg and an older group with 27 heifers aged 9-14 months, weighing 331 ( $\pm$  43) kg. The experimental design was a change-over with two treatments and three three-week periods, where each period consisted of one-week adaptation to the diet and two weeks for registrations of *ad libitum* intake on a group level and recordings of behaviours on ten randomly assigned individuals in each group. The treatments were type of silage and age category of heifers (young and old). Early cut (40% dry matter (DM), 510 g NDF kg<sup>-1</sup> DM and 11.6 MJ metabolizable energy) grass silage with a PS of 231 ( $\pm$  147) mm

was compared with a late cut (48% DM, 643 g NDF kg<sup>-1</sup> DM and 10.0 MJ metabolizable energy) grass silage with a PS of 373 ( $\pm$  245) mm. Early cut, short silage was fed to the younger heifers for two periods and to the older heifers for one period. Trial 2 was conducted at Uddetorp agricultural high school in Skara. Forty-two heifers of the breeds Swedish Red and SF were housed in eight slatted-floor pens, with 22 heifers, aged 5-10 months, weighing 275 ( $\pm$  51) kg, in four pens and 20 heifers, aged 11-16 months, weighing 395 ( $\pm$  72) kg in four pens. The experimental design was a change-over with two treatments and two three-week periods, where each period consisted of one week adaptation to the diet and two weeks for registrations of *ad libitum* intake on a group level and recordings of behaviours on all the individuals in each group. The treatments were type of silage and age category of heifers (young and old). Grass silages (58% vs. 31% DM, 536 vs. 540 g NDF kg<sup>-1</sup> DM and 11.1 vs. 10.2 MJ metabolizable energy) with a PS of 26 ( $\pm$  8.3) mm vs. 254 ( $\pm$  40) mm were compared. For both trials, the silage PS was determined manually from two daily samples during each of the last two weeks in each period. A mixture of the two silages was fed to all heifers during one week prior to start of the trials and for one week between the two periods in trial 2. Heifers within the same 50-kg weight interval were fed equal amounts of concentrates in both trials. The heifers were weighed and scored for body condition at the start and at the end of each three-week period. Direct behavioural observations of several normal and abnormal behaviours were conducted during two (morning and afternoon) and three (morning, noon and afternoon) two-hour sessions in trial 1 and 2, respectively, during the last two days of the last two weeks in each period. Behavioural data from the direct observations was analysed using the generalized linear model procedure (PROC GENMOD) in SAS<sup>®</sup> (Statistical Analysis Systems, version 9.1). Differences between means were declared significant at  $P < 0.05$  and tended to be significant at  $0.05 < P < 0.10$ .

## Results and discussion

Young and old heifers in trial 1 consumed daily 5.4 ( $\pm$  0.8) and 12.6 ( $\pm$  4.8) kg DM, respectively, of the early cut, short silage but only 3.9 ( $\pm$  1.8) and 7.6 ( $\pm$  3.0) kg DM, respectively, of the late cut, long silage. In trial 2, young and old heifers consumed 7.9 ( $\pm$  1.3) and 9.1 ( $\pm$  0.5) kg DM, respectively, of the short silage but 5.6 ( $\pm$  1.2) and 8.0 ( $\pm$  1.6) kg DM, respectively, of the long silage daily. Differences in intake in trial 2 was probably also related to differences in DM and metabolisable energy contents between short and long silages (Krizsan and Randby, 2007). Average daily weight gains of the heifers were 773 ( $\pm$  261) g for the early cut, short silage and 703 ( $\pm$  228) g for the late cut, long silage in trial 1. In trial 2, average daily weight gains of the heifers were 893 ( $\pm$  259) g for short silage and 800 ( $\pm$  269) g for long silage. In trial 1, the young heifers spent more time eating when fed the late cut, long silage compared to when they were fed the early cut, short silage (37.7% vs. 22.4% of observations,  $P < 0.001$ ) but no differences between silage treatments were found for the older heifers (27.7% vs. 25.8% of observations), indicating that the young heifers do not eat long silage as efficiently as the older heifers (Bae *et al.*, 1983). When the heifers in trial 2 were fed short silage, they spent less time eating compared to when they were fed the long silage (Table 1). Young heifers in trial 1 ruminated more when fed the early cut, short silage compared to the late cut, long silage (28.6 vs. 20.1% of observations,  $P < 0.01$ ) whereas no differences between silage treatments were found for the older heifers (18.7% vs. 25.0% of observations). In trial 2, heifers fed the short silage spent more time ruminating compared to heifers fed the long silage (Table 1). The longer rumination time can be related to the higher feed intake when the heifers were fed the early cut and short silages compared to the late cut and long silages. When the heifers were fed the early cut and short silages they were standing idle and lying without ruminating more in both trials than when they were fed the late cut and long silages (Table 1). Consequently, heifers fed the early cut and short silages had more time

available for performing abnormal oral behaviours (Table 1). In both trials, the heifers spent more time licking the fixture when they were fed the early cut and short silages compared to the late cut and long silages indicating that the early cut and short silages do not sufficiently satisfy the heifers' behavioural needs for manipulating the feed (Table 1).

Table 1. Proportion (as percentage) of animals performing the given behaviour at any time during the observation period in relation to treatment.

	Trial 1			Trial 2		
	Early cut short silage	cut long silage	<i>P</i> - value	Short silage	Long silage	<i>P</i> - value
Eating	23.5	31.0	< 0.01	20.3	36.0	< 0.01
Ruminating standing	2.5	3.3	< 0.1	5.8	4.1	< 0.05
Ruminating lying	22.8	20.2	ns	27.5	26.1	< 0.01
Idling, standing	-	-	-	14.9	12.1	< 0.01
Idling, lying	22.5	14.7	< 0.05	22.0	14.7	< 0.01
Lick fixture	0.8	0.5	< 0.05	1.6	0.7	< 0.01

There was no difference between the young and older heifers in the proportion of time they were eating silage. However, the young heifers ruminated more while lying compared to the older heifers in trial 2 (30.4% vs. 22.9% of observations,  $P < 0.05$ ) whereas the older heifers ruminated more while standing compared to the young heifers (trial 1: 4.3% vs. 1.5% of observations,  $P < 0.05$ ; trial 2: 8.0% vs. 2.2% of observations,  $P < 0.10$ ). The young and older heifers in both trials were lying idle and licking the fixture at the same proportion of time.

## Conclusions

Increased silage structure allows the heifers to spend a larger proportion of the day eating compared to a low structure and, thereby, decreases the time available for the heifers to develop and perform abnormal oral behaviours. However, the percentages of observations for abnormal behaviours were less than 2% for both treatments and did not affect the performance of the heifers. Consequently, this has no or very little effect in practice.

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# Milk production systems to increase competitiveness in regions of high rainfall and heavy clay soil types

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## Abstract

The objective of this study was to determine the biological efficiency of two different production systems on a high rainfall heavy clay soil, based on differences in concentrate supplementation levels and stocking rates. Fifty-six spring calving dairy cows were randomized and assigned to a two-treatment (n = 28) feeding system study. Mean lactation number was 1.7, mean calving date 22 February. The two treatments were: low input (LI) and high input (HI) feeding systems. LI had a low stocking rate (2.0 cows ha<sup>-1</sup>), 210 kg N ha<sup>-1</sup>, target of 0.50 Mg concentrate. HI had a stocking rate of 2.7 cows ha<sup>-1</sup>, target concentrate input of 1.5 Mg cow<sup>-1</sup>, and 240 kg N ha<sup>-1</sup>. Total concentrate input was 1.4 Mg concentrate for the HI system and 0.6 Mg for the LI system. Total silage conserved for the two systems was 1.9 Mg dry matter (DM) conserved cow<sup>-1</sup> (LI) and 1.4 Mg DM silage for the HI system. The high concentrate feeding system had a significantly higher (5,236 vs. 4,583 kg cow<sup>-1</sup> P < 0.001) milk yield, solids corrected yield and milk solids production. The mean milk production response to the extra concentrate DM offered was 0.96 kg SCM kg<sup>-1</sup> concentrate DM. Milk output was 14,137 kg ha<sup>-1</sup> for the HI system compared with 9,624 kg ha<sup>-1</sup> for LI system. To increase competitiveness in both systems grazing days must be optimized. Dependency on grass silage in such regions can be reduced with a more flexible approach to grazing management particularly in spring and autumn.

Keywords: milk production, grass, dairy cows

## Introduction

There is a large variation in the cost of milk production in specialized dairy farms in Ireland (Dillon et al., 2007). Some of the variation in cost may be associated with variation in soil type and climatic conditions. In Ireland, the two factors that have largest influence on animal production from grassland farming are soil type and climate conditions. The results of previous work at the Kilmaley Research Farm indicate that the most profitable spring milk production system is based on a feed budget of 2.1 Mg of grass silage, 2.8 Mg of grazed grass and 0.5 Mg of concentrates per cow at a stocking rate of 2.0 cows ha<sup>-1</sup> with a nitrogen input of 210 kg ha<sup>-1</sup>. An alternative system of milk production for high rainfall, heavy clay soils is a higher concentrate feeding system allowing for higher animal performance per unit area. This system will be less dependent on achieving high animal performance from grass silage, while at the same time lowering fixed costs per unit of output. If concentrate supplementation could be used efficiently, allowing a higher stocking rate to be carried on the farm, this could potentially result in profits similar to pasture-based systems in free-draining soils with low rainfall. The objective of this study was to determine the biological efficiency of two different production systems on a high rainfall, heavy clay soil based on differences in concentrate supplementation levels and stocking rates.

## Materials and methods

The experiment commenced in February 2006 and was completed in December 2006. Fifty-six spring calving dairy cows were randomized and assigned to a two-treatment ( $n = 28$ ) feeding system study, 0.8 of the herd were primiparous. Mean lactation number was 1.7 (s.d. 1.8), mean calving date 22 February (s.d. 18 days), respectively.

The two treatments were: low input (LI) and high input (HI). LI had a low stocking rate of 2.0 cows  $\text{ha}^{-1}$ , 210 kg N  $\text{ha}^{-1}$ , a target 500 kg concentrate, and 2 main silage cuts. The high concentrate system (HI) had a stocking rate of 2.7 cows  $\text{ha}^{-1}$ , a target concentrate input of 1.5 Mg  $\text{cow}^{-1}$ , 240 kg N  $\text{ha}^{-1}$  and one main silage cut.

Separate farmlets, each of 19 individual paddocks, were in operation for both systems. In both systems from turnout the objective was to minimize grass silage feeding. Once grass supply was sufficient the LI herd was managed on a grass-only diet, while the HI herd was supplemented with 4 kg concentrate throughout the grazing season. A constant differential of 4 kg concentrate was maintained between the herds throughout the season (when concentrate was offered to the LI herd). Turnout was in late March with housing by night in late October. Milk yield was recorded daily, and milk composition and bodyweight were recorded weekly. All animal variables were analysed using covariate analysis in SAS (2006).

## Results and discussion

Mean grazing season length was 167 full time grazing days, which was 209 days at grass for both systems, with grazing beginning in late February and finishing in early November; full-time grazing was suspended a number of occasions during the season due to adverse weather conditions (notable months were April, May and October). Total concentrate input was 1.395 Mg concentrate for the HI system, and 0.615 Mg for the LI system. Total silage conserved for the two systems was 1.54 Mg dry matter (DM) conserved  $\text{cow}^{-1}$  (LI), and 1.2 Mg DM silage for the HI system. The high concentrate feeding system had a significantly higher ( $P < 0.001$ ) milk yield, solids corrected yield and milk solids production (Table 1). The mean milk production response to the extra concentrate DM offered was 0.96 kg SCM  $\text{kg}^{-1}$  concentrate DM. Feeding system had no significant effect on milk composition, bodyweight or body condition score (BCS). There were no significant differences between systems for minimum bodyweight or BCS. Milk output was 14,137 kg for the HI system compared with 9,624 kg  $\text{ha}^{-1}$  for the LI system. This is likely to be below potential of the two systems given the high percentage of primiparous animals lactating in this year of the study.

Table 1. Effect of feeding system on total lactation milk production performance.

	LI	HI	Sed	Sig
Milk yield (kg)	4583	5236	205.6	***
Solids corrected milk (kg)	4189	4843	189.4	***
Milk fat (g $\text{kg}^{-1}$ )	39.2	40.1	0.124	ns
Milk protein (g $\text{kg}^{-1}$ )	32.0	32.3	0.044	ns
Milk lactose (g $\text{kg}^{-1}$ )	45.9	46.1	0.039	ns
Milk solids (kg)	327	377	15.16	***
Bodyweight (kg)	488	486	12.9	ns
Body condition score	2.92	2.90	0.063	ns

Significance; \*\*\*  $P < 0.001$ ; ns = Non-significant; sed – standard error of difference.

## Conclusions

Grazing days were below, and concentrate input above, the targets for both systems in this year of the study. To increase competitiveness in both systems grazing days must be



optimized. Dependency on grass silage in such regions (high rainfall, heavy soil) can be reduced with a more flexible approach to grazing management, particularly in spring and autumn. To maintain a viable milk production enterprise in such regions, target milk output per ha must be comparable to that achieved from drier soil types (Horan *et al.*, 2005).

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# Are dairy cow rumen pH levels affected by herbage mass and daily herbage allowance?

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## Abstract

The objective of this study was to characterize the rumen pH of dairy cows grazing ryegrass pastures with two different levels of pre-grazing herbage mass and daily herbage allowance (DHA). Eight cannulated cows were allocated to four grazing treatments. High and low herbage mass (2,200 and 1,500 kg DM ha<sup>-1</sup> respectively), at two DHA (16 and 20 kg cow<sup>-1</sup> day<sup>-1</sup>) were compared. Rumen pH was measured at four periods (between June and August) at 07.00, 11.00, 15.00 and 20.00 h. Milk production and composition were collected weekly during each period as well as herbage measurements. The mean rumen pH for all the treatments was 5.87 with a maximum of 7.00 and a minimum of 5.35. Rumen pH tended to be lower at low herbage mass (5.82 vs. 5.88 in the high herbage mass treatment;  $P = 0.09$ ) and was significantly different for the treatment low herbage mass and low DHA (pH = 5.77;  $P < 0.05$ ). Milk yield was higher for high DHA. In conclusion, sward characteristics (herbage mass) affect rumen pH levels and cows grazing such swards had low rumen pH levels ( $< 6.00$ ) for long periods of the day.

Keywords: rumen pH, daily herbage allowance, herbage mass

## Introduction

It is commonly accepted that a rumen pH under 6.0-6.2 affects fibre digestion (Hoover *et al.*, 1986; Pitt *et al.*, 1996). In grazing systems, this scenario may affect the production because forage is the main feed. Several authors indicated that in grazing systems, cows present pH variations between 5.6-6.8 (Holden *et al.*, 1994; Carruthers and Neil, 1997; Carruthers *et al.*, 1997). Taweel *et al.* (2005) observed an average pH of 5.8 in cows grazing perennial ryegrass plus 4.6 kg of concentrate (as fed). When cows are grazing at different allowances, the rumen pH can be affected as well (Stockdale *et al.*, 2005). The aim of this work was to characterize the rumen pH of dairy cows grazing ryegrass pastures at two levels of pre-grazing herbage mass and two levels of daily herbage allowance. Also, effects on milk production and milk composition are discussed.

## Materials and methods

The experiment was carried out in Moorepark Research Centre, Fermoy, Co. Cork, Ireland (50°7'N; 8°16'W). A perennial ryegrass pasture was used. Eight cannulated cows were allocated to four grazing treatments in two 4 x 4 Latin square designs and data were collected during 4 periods (a week period) from June to August of 2007. The treatments were high and low herbage mass (2,200 and 1,500 kg DM ha<sup>-1</sup>), at two levels of daily herbage mass (DHA, 16 and 20 kg DM cow<sup>-1</sup> day<sup>-1</sup>). Rumen pH was measured during two consecutive days at 07.00, 11.00, 15.00 and 20.00 h in each period. Milk production was recorded daily during each period and milk fat, protein and lactose were determined from one successive a.m. and p.m. sample taken during the week. Herbage mass ( $> 4$  cm) was determined twice weekly by cutting two strips per paddock, and then ten grass height measurements were recorded before



and after harvesting in each cut strip using an electronic plate meter. This allowed the calculation of the sward density ( $\text{kg of DM cm}^{-1} \text{ ha}^{-1}$ ). Pre- and post-grazing sward heights were calculated using the rising plate meter 35 times per paddock across the two diagonals. The measured pre-grazing sward height, multiplied by the sward density, was used to calculate the DHA. Data were analysed by ANOVA using PROC MIXED (SAS Institute, 2002). Rumen pH, milk production and milk composition were analysed using the following models with the variable 'cow' as random effect:

#### Rumen pH

$$Y_{ijkl} = \mu + P_i + T_j + A_k + H_l + A_k * H_l + T_j * A_k * H_l + P_i * A_k * H_l + e_{ijkl}$$

#### Milk yield and composition

$$Y_{ijk} = \mu + P_i + A_j + H_k + P_i * A_j + P_i * H_k + A_j * H_k + P_i * A_j * H_k + e_{ijk}$$

where Y represents the response,  $\mu$  is the mean, P is the period, T is the time (07.00, 11.00, 15.00 and 20.00 h), A is the DHA treatment and H is the pre-yield herbage mass treatment. Tukey test was performed for multiple comparison and significant differences were declared at  $P < 0.05$  and trends were declared at  $P < 0.10$ .

### Results and discussion

The herbage mass values were  $2,580 \pm 517.1$  and  $1,726 \pm 303.6$  kg of DM  $\text{ha}^{-1}$  for high and low herbage mass treatments, respectively. The DHA was adjusted according to the herbage mass and were 16 and 20 kg of DM  $\text{cow}^{-1} \text{ day}^{-1}$  for high and low DHA treatments. The mean rumen pH for all the treatments was 5.87, with a maximum of 7.00 and a minimum of 5.35. Rumen pH tended to be lower at low herbage mass (5.82 vs. 5.88 in the high herbage mass treatment;  $P = 0.09$ ) and was significantly different for the treatment low herbage mass and low DHA (pH = 5.77; Table 1). In this work, the low herbage mass treatment had a better quality of sward in comparison with the high herbage mass. It is possible that cows grazing a better sward present lower pH, associated with a less intake of effective fibre and a greater production of volatile fatty acids (VFA). The combination of low herbage mass with low DHA presented the lowest pH. This effect may be explained by the previous comment and by the finding of Wales *et al.* (2001), who found that cows grazing perennial ryegrass at low DHA ruminated less, although the authors did not find differences in rumen pH. Contrary to this, Stockdale *et al.* (2005) found that cows grazing Persian clover or perennial ryegrass at low herbage allowance presented a higher rumen pH. Finally, rumen pH was affected by time ( $P < 0.01$ ) decreasing during the day (all treatments dropped from pH 6.33 at 07.00 h to 5.55 at 20.00 h) and by period ( $P < 0.01$ ), being the first period which presented the highest pH values (data not shown).

Milk yield, fat and protein content were not different for high or low herbage mass. However, milk yield was significantly higher ( $P < 0.05$ ) for high DHA (Table 1) probably as consequence of a highest intake. Cows grazing the low DHA treatment recorded the lowest milk yield (0.8 kg  $\text{day}^{-1}$  less). Milk protein was numerically higher with the higher DHA treatments.

Table 1. Milk yield, milk composition and rumen pH.

Variable	Treatments					Sig		
	High Herbage mass		Low Herbage mass		RSE	Herbage mass	DHA	Herbage mass x DHA
	High	Low	High	Low				
	DHA	DHA	DHA	DHA				
Milk yield (kg d <sup>-1</sup> )	20.8	20.1	21.1	20.0	1.83	ns	*	ns
Fat (g kg <sup>-1</sup> )	38.5	40.1	39.4	40.5	0.37	ns	ns	ns
Protein (g kg <sup>-1</sup> )	34.1	33.7	34.4	33.7	0.20	ns	ns	ns
pH	5.85a	5.90a	5.87a	5.77b	0.032	ns	ns	*

## Conclusions

Cows grazing all swards had low rumen pH levels ( $< 6.00$ ) for long periods of the day. The low herbage mass, low DHA treatment recorded the lowest pH level, and pH tended to be lower at low herbage mass. It appears that grazing dairy cows can allow rumen pH levels to drop to  $< 6.0$  during the grazing day. It appears that offering a low DHA with a leafy sward (low herbage mass) may reduce rumen pH levels to periodically low levels. However, these low rumen pH levels had little effect on milk production performance. Effects of supplementation with a source of effective fibre should be studied in future work.

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# Effect of harvesting time and wilting of silage on digestibility in cows and sheep

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## Abstract

Round bale grass silage harvested at early (D-value 757 g kg<sup>-1</sup> DM) or normal (D-value 696 g kg<sup>-1</sup> DM) time, direct cut or wilted, was used to study effect of harvesting time and wilting on digestibility of main nutrients. Fermentation quality was good in all 4 silages. Silages were fed *ad libitum* to 8 cows (Norwegian Red) in early lactation. Animals were split by two concentrate levels (4 and 10 kg d<sup>-1</sup>) and used in 4 x 4 Latin square experiments (4-week periods). No major effects of wilting on digestibility of nutrients were observed. Two weeks delayed harvesting time reduced dietary organic matter digestibility in cows from 813 to 759 g kg<sup>-1</sup> DM at low concentrate level, and from 776 to 748 g kg<sup>-1</sup> DM at high concentrate level. Corresponding values for NDF digestibility were 788 and 724, and 702 and 626 g kg<sup>-1</sup> DM at low and high concentrate level, respectively. Energy supply calculated on the basis of digestibility measures of silage and concentrate with sheep agreed well with the calculation from ration digestibility in cows at 4 kg concentrate level. At 10 kg concentrate level, calculated energy supply was higher based on sheep than on cows.

Keywords: silage, harvesting time, wilting, digestibility

## Introduction

High digestibility, especially of fibre, is critical for the intake capacity of silage (Huhtanen *et al.*, 2007) and this largely explains increased intake of early, compared with 2-weeks later harvested silage, found by Prestløkken *et al.* (2008). In this paper, ration digestibility of main nutrients obtained with 8 cows in that experiment is presented, and also digestibility of the same nutrients in silages and concentrates measured with sheep. The relation between these two measures of digestibility and the influence on calculated energy value is discussed.

## Materials and methods

Silage quality, feeding management, sampling and experimental design were as presented by Prestløkken *et al.* (2008). Of the 32 cows used in the experiment, total ration digestibility was measured on 8 cows using total collection of faeces over a 72 h period. Four cows received 4 kg of a protein concentrate, whereas 4 other cows received additionally 6 kg of a cereal concentrate, giving a total of 10 kg concentrate. Each concentrate level constituted a Latin square design including four silages. Digestibility of concentrates and silages was determined using castrated wethers at maintenance feeding. Energy values were calculated according to Ekern *et al.* (1991). Metabolizable energy (ME) in the cow diet was calculated using digestibility on the actual feed intakes, and ME equations given by Ekern *et al.* (1991).

## Results and discussion

Chemical composition of silage and concentrate is shown in Table 1. Wilting increased dry matter content in silage marginally. Fermentation quality was good in all silages making them well suited for studying the effect of harvesting time and wilting on digestibility of nutrients.

Table 1. Chemical composition (g kg<sup>-1</sup> DM) in experimental feeds.

	Silage quality <sup>1</sup>				Response <sup>2</sup>		Concentrate <sup>3</sup>	
	ED	EW	ND	NW	Early	Wilting	Cereal	Protein
Dry matter, g kg <sup>-1</sup>	187	235	202	233	- 6	40	870	872
Organic matter	930	927	939	932	-7	- 5	955	904
Protein	202	181	138	139	54	-10	135	275
NDF	450	447	545	548	-98	0	190	180

<sup>1</sup>ED = Early direct, EW = early wilted, ND = Normal direct, NW = Normal wilted. <sup>2</sup>Response: Early = early minus normal. Wilting = wilted minus direct. <sup>3</sup>For composition see Prestløkken *et al.* (2008).

Apparent digestibility of dry matter, organic matter, protein and NDF in the silage determined with sheep is shown in Table 2. The values for the concentrates are shown in Table 3. No effect of wilting on digestibility was observed. Increased maturity stage reduced digestibility of all nutrients, as expected. The reduction in organic matter digestibility by a 2-week delay in harvesting time was *ca.* 5 g d<sup>-1</sup>, which was somewhat higher than reported by Rinne *et al.* (1999) (*ca.* 3 g d<sup>-1</sup>). An energy value close to 1 FEm kg<sup>-1</sup> DM is high for grass silage.

Table 2. Digestibility of silages determined with sheep, and calculated energy value.

	Silage quality <sup>1</sup>				SEM	Response <sup>2</sup>		Effects <sup>3</sup>		
	ED	EW	ND	NW		Early	Wilting	E	W	E*W
<u>Digestibility:</u>										
Dry matter	.796	.798	.728	.726	.001	.071	.000	***		
Organic matter	.814	.816	.747	.741	.008	.071	-.002	***		
Protein	.785	.802	.705	.676	.005	.103	-.006	***		**
NDF	.833	.825	.758	.744	.010	.078	-.010	***		
<u>Energy value<sup>4</sup>:</u>										
ME, MJ kg <sup>-1</sup> DM	11.6	11.5	10.5	10.3	0.1	1.2	-0.2	***		
FEm, kg <sup>-1</sup> DM	0.99	0.99	0.88	0.87	0.01	0.12	-0.01	***		

<sup>1</sup>See Table 1. <sup>2</sup>See Table 1. <sup>3</sup>Effects of: harvesting time (E), wilting (W) and interaction (E\*W). Significance: \* =  $P < 0.05$ , \*\* =  $P < 0.01$ , \*\*\* =  $P < 0.001$ . Tendency is given with number. <sup>4</sup>Calculated according to Ekern *et al.* (1991). SEM = Standard Error Means. ME = Metabolizable energy, FEm = Feed Units milk.

Table 3. Digestibility of concentrates determined with sheep, and calculated energy value<sup>1</sup>.

	Digestibility				Energy value, kg <sup>-1</sup> DM	
	DM	OM	Protein	NDF	ME, MJ	FEm
Cereal compound	.815	.828	.753	.455	13.1	1.18
Protein compound	.824	.874	.889	.634	13.9	1.27

<sup>1</sup>Calculated according to Ekern *et al.* (1991).

No effect of wilting on ration digestibility was observed, whereas the effect of harvesting time was considerable (Table 4). The high digestibility observed with cows fed early harvested silage with 4 kg concentrates indicates that the digestive tract had worked properly with those high-forage rations. However, although silage intake, milk yield and body weight gain were numerically highest when early-cut wilted silage was fed, this ration was also most over predicted in energy value. The ration FEm value tended to be higher at 4 than 10 kg concentrate level at early, but not at normal harvesting time. This indicates that the increased concentrate level influenced nutrient digestibility negatively, probably fibre. The reduction in NDF digestibility was *ca.* 100 g kg<sup>-1</sup> DM, of which only 30-40 g kg<sup>-1</sup> DM can be attributed to low digestible NDF from concentrate. In line with this, use of cow total ration digestibility predicted the energy value of the feed rations better than digestibility trials with sheep. The difference between cow and sheep digestibility was greater at 10 than 4 kg concentrate, probably because sheep did not express reduced fibre digestibility in high concentrate rations. At 10 kg concentrate, energy supply based on ration digestibility predicted an undersupply of energy at normal harvesting time. Further calculations on energy utilization are needed.

Table 4. Feed intake, milk production and energy calculations at 4 and 10 kg concentrate.

	Silage quality <sup>1</sup>				SEM	Response <sup>2</sup>		Effects <sup>3</sup>		
	ED	EW	ND	NW		Early	Wilting	E	W	E*W
<b>4 kg Concentrate;</b>										
<u>Feed intake, milk production and weight change, d<sup>-1</sup>:</u>										
Silage, kg DM	16.4	19.2	15.1	15.1	0.7	2.7	1.4	**	0.09	
Concentrate, kg DM	3.5	3.5	3.5	3.5						
Energy corrected milk, kg	28.5	30.0	25.6	23.3	1.9	4.8	-0.4	*		
Weight change, kg	.089	.286	.232	.161	.189	-.009	.063			
<u>Digestibility:</u>										
Dry matter	.802	.802	.745	.752	.012	.053	.004	**		
Organic matter	.812	.814	.757	.761	.012	.053	.003	**		
Protein	.790	.771	.735	.750	.010	.037	-.002	**		
NDF	.789	.786	.723	.724	.016	.064	-.001	**		
<u>Energy value<sup>4</sup>, kg<sup>-1</sup> DM:</u>										
ME, MJ	11.7	11.6	10.8	10.8	0.16	0.9	-0.1	**		
FEm	1.01	1.00	0.91	0.92	0.02	0.10	0.00	**		
<u>Energy supply<sup>4</sup> (FEm), d<sup>-1</sup>:</u>										
Requirement	18.3	19.0	16.8	15.8	0.9	2.4	-0.2	*		
Based on cows	20.1	22.7	17.0	17.0	0.9	4.4	1.3	**		
Based on sheep	20.8	23.6	17.7	17.5	0.7	1.3	4.6	***		0.09
<b>10 kg Concentrate;</b>										
<u>Feed intake, milk production and weight change, d<sup>-1</sup>:</u>										
Silage, kg DM	11.7	13.8	11.3	10.7	0.5	1.8	0.8	*		*
Concentrate, kg DM	8.0	7.6	8.7	8.7	0.4	-0.9	-0.2	*		
Energy corrected milk, kg	30.2	31.7	30.9	30.1	0.9	0.5	0.4			
Weight change, kg	.259	.259	.018	.313	.198	.094	.148			
<u>Digestibility:</u>										
Dry matter	.762	.788	.741	.734	.007	.038	.010	**		0.05
Organic matter	.773	.799	.752	.744	.007	.038	.009	**		*
Protein	.736	.750	.715	.719	.007	.025	.009	*		
NDF	.679	.725	.633	.619	.018	.076	.016	**		
<u>Energy value<sup>4</sup>, kg<sup>-1</sup> DM:</u>										
ME, MJ	11.3	11.5	10.9	10.8	0.09	0.6	0.1	**		0.08
FEm	0.98	1.01	0.94	0.93	0.01	0.06	0.01	**		0.06
<u>Energy supply<sup>4</sup> (FEm), d<sup>-1</sup>:</u>										
Requirement	19.3	20.1	19.7	19.3	0.4	0.2	0.2			
Based on cows	19.1	21.3	18.7	17.8	0.5	2.1	0.6	**		*
Based on sheep	21.4	23.0	20.6	19.8	0.6	2.0	0.4	*		0.08

<sup>1</sup>See Table 1. <sup>2</sup>See Table 1. <sup>3</sup>See Table 2. <sup>4</sup>Calculated according to Ekern *et al.* (1991).

## Conclusions

Early harvested wet silage was highly digested by dairy cows. Digestibility was considerably reduced in normal compared to early harvested silage. No effect of wilting on digestibility was observed. Energy evaluation based on digestibility in sheep over predicted the energy supply at 10 kg, but not to the same extent at 4 kg concentrate supply.

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# Effect of harvesting time and wilting on feed intake and milk production

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## Abstract

Round bale grass silage harvested early (D-value 757 g kg<sup>-1</sup> DM) or at a normal (D-value 696 g kg<sup>-1</sup> DM) time, direct cut or wilted, was used to study the effect of harvesting time and wilting on intake and production by dairy cows. Fermentation quality was good in all four silages. Silages were fed *ad libitum* to 32 cows (Norwegian Red) in early lactation. Animals were split in groups of two concentrate levels (4 and 10 kg d<sup>-1</sup>) and used in 4 x 4 Latin square experiments (4-week periods). Wilting increased dry matter (DM) in silage slightly from 195 to 234 g kg<sup>-1</sup> DM. Delayed harvesting reduced protein (192 to 139 g kg<sup>-1</sup> DM) and increased NDF (449 to 547 g kg<sup>-1</sup> DM) in silage. Wilting increased DM intake of silage (+2.3 kg DM), and milk yield (+1.1 kg d<sup>-1</sup>) at early, but not at normal harvesting time. Early harvesting increased silage intake at low (+2.1 kg DM) and high (+2.3 kg DM) concentrate level, but the effect on milk yield was higher at low (+3.3 kg d<sup>-1</sup>) than high (+1.1 kg d<sup>-1</sup>) concentrate level.

Keywords: silage, harvesting time, wilting, concentrate

## Introduction

Voluntary intake of digestible nutrients is a main factor determining the production potential of dairy cows. Early harvesting increases the intake potential of grass silage and thus the milk production potential of the cow. However, early harvested grass normally has a low content of dry matter and is not easy to ensile. Wilting increases dry matter content and makes it easier to achieve a good fermentation quality. The objective of this study was to evaluate the effect of harvesting time and wilting of grass silage on feed intake and milk production in cows.

## Materials and methods

A primary growth of a sward of timothy (*Phleum pratense*), meadow fescue (*Festuca pratensis*) and red clover (*Trifolium pratense*) was harvested at early (mean stage by weight (MSW) = 2.44) and normal (MSW = 3.01) (Moore *et al.*, 1991) harvesting time. The crop was direct cut or wilted before it was compressed in a round baler (Orkel GP1260) and wrapped in 6 layers of plastic film. To obtain a uniform fermentation quality, a formic acid based additive (GrasAAT Lacto, 780 g formic acid kg<sup>-1</sup>) was added at 4.5, 4.0, 3.5 and 3.2 l ton<sup>-1</sup> in the direct early cut (ED), wilted early cut (EW), direct normal cut (ND), and wilted normal cut (NW) silages, respectively. The silages were cut to a particle length of 60 to 100 mm and fed *ad libitum* to 32 cows (Norwegian Red). The animals were split into groups by two concentrate levels (4 and 10 kg d<sup>-1</sup>) successively when calving. All cows were offered a standard silage *ad libitum* until the onset of the study. The cows (46 ± 17 DIM, 586 ± 76 kg BW) had a milk yield of 26.1 ± 3.6 and 29.5 ± 2.8 kg d<sup>-1</sup> at the 4 and 10 kg concentrate level, respectively, when the experiment started using balanced 4 x 4 Latin square designs with 4-week periods. The cows were housed in tie-stalls and had access to silage 12 hours daily (0330 to 0800 h, 1200 to 1630 h, and 1900 to 2200 h). Fresh silage was fed twice (1000 and 1630 h), whereas concentrate was fed in four equal meals (0330, 0600, 1200 and 1500 h) daily. All animals received 4 kg d<sup>-1</sup> of a protein compound, whereas the 10 kg group got additionally 6 kg d<sup>-1</sup> of a cereal compound daily. Feed intake was registered 5 days and milk yield 4 days a week. Milk



was analysed with FTIR. Feed was analysed for chemical composition and fermentation quality. Milk taste was measured with a test panel using a scale where 5 was good and 1 was bad milk (Norsk Matanalyse, Oslo).

## Results and discussion

Chemical composition of silage and concentrate, and fermentation quality of silage are shown in Table 1. Wilting increased dry matter in silage by an average of 40 g kg<sup>-1</sup>, which was lower than aimed for due to rain showers. Fermentation quality was good in all silages and not expected to influence voluntary intake of silage negatively (Huhtanen *et al.*, 2002).

Table 1. Chemical composition and fermentation quality of feeds (g kg<sup>-1</sup> DM).

	Silage quality <sup>1</sup>				Response <sup>2</sup>		Concentrate <sup>3</sup>	
	ED	EW	ND	NW	Early	Wilting	Cereal	Protein
Dry matter, g kg <sup>-1</sup>	187	235	202	233	- 6	40	870	872
Organic matter	930	927	939	932	-7	- 5	955	904
Protein	202	181	138	139	54	-10	135	275
Fat	48	43	33	33	12	- 3	55	54
NDF	450	447	545	548	-98	0	190	180
Starch							520	298
NH <sub>3</sub> -N, g kg <sup>-1</sup> N	99	78	94	100	- 9	- 8		
pH	4.14	4.18	3.99	4.07	0.13	0.06		
Lactic acid	66.8	66.7	65.8	61.2	3.2	- 2.3		
Acetic acid	18.9	17.0	23.3	13.9	- 0.7	- 5.6		
Propionic acid	1.98	1.90	1.46	1.34	0.53	- 0.1		
Butyric acid	0.71	0.00	0.07	0.00	0.32	- 0.39		
Ethanol	7.6	9.9	19.6	22.1	-12.1	2.3		

<sup>1</sup>ED = Early direct, EW = early wilted, ND = Normal direct, NW = Normal wilted. <sup>2</sup>Response: Early = early minus normal. Wilting = wilted minus direct. <sup>3</sup>Concentrate (g kg<sup>-1</sup>): Cereals = 607 g barley, 250 g oats, 50 g ExPro 00E (heat treated rapeseed meal), 70 g molasses, 10 g fat, 13 g minerals. Protein = 334 g barley, 100 g oats, 50 g peas, 150 g soybean meal (SBM), 30 g SoyPass (lignosulphonate treated SBM), 150 g ExPro 00E, 30 g corn gluten meal, 30 g fish meal, 70 g molasses, 10 g fat, 46 g minerals and vitamins.

On average, early harvesting increased silage intake by 2.1 kg DM at the 4 kg concentrate level, and 2.3 kg DM at the 10 kg concentrate level, giving intakes of 17.1 and 13.7 kg DM d<sup>-1</sup>, respectively for early, and 15.1 and 11.4 kg DM d<sup>-1</sup> for normal harvested silage (Table 2). Wilting increased silage intake at early, but not at normal harvesting time. The effect was more pronounced at 4 (+ 2.3 kg d<sup>-1</sup>) than 10 (+ 1.8 kg d<sup>-1</sup>) kg concentrate level. In line with this, wilting tended to increase milk yield (+ 1.1 kg d<sup>-1</sup>) at early, but not at normal harvesting time, and only at the low concentrate level. Also, previous studies have shown small, if any, yield responses to silage wilting when the direct-cut control silage is well fermented, i.e. when effective silage additives were applied to direct-cut silage (Zimmer and Wilkins, 1984).

Ration dry matter intake was higher at early harvesting and 4 kg concentrate than at normal harvesting and 10 kg concentrate supplementation. Moreover, early harvesting increased milk yield, but the effect was higher at low (+ 3.3 kg d<sup>-1</sup>) than at high (+ 1.1 kg d<sup>-1</sup>) concentrate level (Table 2). This confirms that early harvesting is more important at low than high concentrate supplementation (Kristensen and Skovborg, 1990). Early harvesting increased milk protein content both at 4 and 10 kg concentrate levels, indicating a better energy status of the animals.

Especially at low concentrate level, early harvested wilted silage increased the incidence of off-flavour in the milk. Transmitted off-flavour (Shipe *et al.*, 1978) was dominant, indicating that the problem is feed related. The increase in off-flavour cannot be explained by increased level of free fatty acids in the milk, or more ethanol in the silage, as both these parameters were lowest at earliest harvesting. The utilization of dietary N into milk was lowest at early



harvesting and low concentrate level, and highest at normal harvesting and high concentrate level. Thus, harvesting time and concentrate supplementation affects the dietary N utilization.

Table 2. Effect of silage quality on feed intake, and production of milk and energy corrected milk (ECM), milk constituents, free fatty acids (FFA), urea, milk taste, and utilization of N.

	Silage quality <sup>1</sup>				SEM	Response <sup>2</sup>		Effects <sup>3</sup>		
	ED	EW	ND	NW		Early	Wilting	E	W	E*W
<u>4 kg concentrate:</u>										
Silage, kg DM	15.9	18.2	15.1	15.0	0.43	2.1	1.1	***	*	**
Concentrate, kg DM	3.5	3.5	3.5	3.5	0.01	0.0	0.0			
NDF, g kg <sup>-1</sup> BW	13.4	14.9	15.0	14.8	0.38	0.7	0.7	***	0.07	*
Milk, kg	27.9	29.0	25.5	24.8	0.51	3.2	0.2	***		0.08
ECM, kg	27.7	29.3	25.3	25.0	0.46	3.3	0.6	***		0.06
Fat, g kg <sup>-1</sup>	39.9	40.9	40.0	41.1	0.6	-0.2	1.0		0.07	
Protein, g kg <sup>-1</sup>	32.5	32.8	32.1	32.3	0.2	0.4	0.2	0.06		
Lactose, g kg <sup>-1</sup>	46.8	46.9	46.8	46.8	0.1	0.0	0.0			
FFA, meq L <sup>-1</sup>	0.73	0.69	1.01	0.90	0.04	-0.24	-0.08	***	*	
Urea, mM	6.87	6.28	5.68	5.93	0.22	0.77	-0.18	**		0.06
Taste	3.90	2.98	4.00	4.01	0.16	-0.57	-0.46	***	**	***
N utilization	0.206	0.212	0.258	0.254	0.006	-0.048	0.001	***		
<u>10 kg concentrate:</u>										
Silage, kg DM	12.8	14.6	11.7	11.0	0.25	2.3	0.5	***	**	***
Concentrate, kg DM	8.4	8.1	8.6	8.7	0.12	-0.3	0.2	**	0.09	
NDF, g kg <sup>-1</sup> BW	12.1	13.0	13.0	12.4	0.24	-0.2	0.2	***		**
Milk, kg	31.3	30.8	30.1	29.7	0.41	1.1	-0.4	**		
ECM, kg	31.1	31.0	30.0	29.7	0.44	1.2	-0.2	**		
Fat, g kg <sup>-1</sup>	38.7	39.6	39.4	39.4	0.4	-0.3	0.4			
Protein, g kg <sup>-1</sup>	33.8	33.9	32.7	33.1	0.2	0.9	0.2	***		
Lactose, g kg <sup>-1</sup>	48.4	48.5	48.8	48.6	0.1	-0.2	0.0	*		
FFA, meq L <sup>-1</sup>	0.52	0.52	0.64	0.65	0.02	-0.12	0.00	***		
Urea, mM	5.49	5.27	4.65	4.77	0.09	0.67	-0.05	***		0.06
Taste	3.75	3.49	3.80	4.13	0.14	-0.35	0.04	**		**
N utilization	0.239	0.237	0.288	0.296	0.004	-0.054	0.003	***		

<sup>1</sup>See Table 1. <sup>2</sup>See Table 1. <sup>3</sup>Effect of: harvesting time (E), wilting (W) and interaction (E\*W).

Significance: \* = P < 0.05, \*\* = P < 0.01, \*\*\* = P < 0.001. Tendency is given with number. SEM = Standard Error Mean. Milk taste on a scale were 5 is good and 1 is bad milk. N utilization as dietary N in milk.

## Conclusions

The intake potential for early harvested grass silage is high. The response to concentrate supplementation was low for cows fed early harvested silage. Thus, early harvesting has a potential for high feed intake and milk production at low concentrate supplementation. The effect of wilting was in general small. Utilization of N to milk needs to be improved.

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# Performance of dairy steers fed whole-crop barley silages harvested at different stages of maturity

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## Abstract

Sixty-three younger (Exp. 1, live weight (LW) 119 kg) and 48 older (Exp. 2, LW 414 kg) pen-housed dairy steers were used in twelve-week long trials to investigate effects of maturity stage of whole-crop barley silages (WCBS) on animal performance. Precision-chopped WCBS harvested at heading (H), milk- (M) and dough stage (D) of maturity were fed *ad libitum* and supplemented with concentrates at 0.6-1 kg head<sup>-1</sup> d<sup>-1</sup>. Feed intake and LW were registered continuously and data were analysed by ANOVA using GLM. Silage DM intake was highest for D in both experiments; 20.5<sup>a</sup> vs. 19.1<sup>b</sup> and 17.7<sup>c</sup> g kg<sup>-1</sup> LW d<sup>-1</sup> in Exp. 1 and 20.2<sup>a</sup> vs. 17.5<sup>b</sup> and 17.1<sup>b</sup> g kg<sup>-1</sup> LW d<sup>-1</sup> in Exp. 2 for D, M and H silages, respectively ( $P < 0.001$ ). Feed conversion, expressed as kg silage DM intake kg<sup>-1</sup> LW gain, was lowest for H in Exp. 1; 3.7<sup>b</sup> vs. 4.4<sup>a</sup> and 4.3<sup>a</sup> kg kg<sup>-1</sup> and lower than M in Exp 2; 8.6<sup>b</sup> vs. 11.8<sup>a</sup> and 10.7<sup>ab</sup> kg kg<sup>-1</sup> for H, M and D silages, respectively ( $P < 0.05$ ). The results indicate that WCBS should be harvested at heading or dough stage for good animal performance.

Keywords: whole-crop cereal silage, dairy steers, feed intake

## Introduction

Maturity stage at harvest of whole-crop cereals affects digestibility and crop yield and is thereby a crucial factor in the production and utilization of whole-crop cereal silage (Baron *et al.*, 1992; Südekum and Arndt, 1998). Whole-crop cereal silage is often harvested at late maturity stages when yield is high but could also be harvested at earlier maturity stages for several reasons. When whole-crop cereals are used as nurse crops for establishing leys, harvest at early maturity stages will increase the time for irradiation to the undersown ley and thereby improve the vigour of the sward with possibilities for an autumn harvest during the establishment year. At early maturity stages, the moisture content, as well as the sugar content, is higher than at later maturity stages, which facilitates the fermentation process during ensiling (Nadeau, 2007). As digestibility and chemical composition vary with maturity of whole-crop cereals, maturity stage may also affect feed intake, which has a significant impact on animal performance. The aim of this study was to evaluate the effect of maturity stage at harvest of whole-crop barley silage on feed intake and live weight gain in growing dairy steers.

## Materials and methods

Spring barley (cv. Kinnan) was sown on April 4, 2003 and harvested at heading on June 26 (code 59 according to Zadoks *et al.*, 1974), at early milk stage on July 7 (code 73) and at mid-dough stage on July 21 (code 85) at Götala Research Station, Skara, Sweden. Crops were cut with a mower conditioner, precision chopped to 20 mm theoretical chopping length and stored in bunker silos covered with plastic and sand on top. Four litres of Kofasil® Majs (sodium propionate and sodium benzoate, Addcon Agrar GmbH, Bonn, Germany) Mg<sup>-1</sup> fresh matter was applied with a pump and sprayer system at chopping.

In Experiment (Exp.) 1, 63 dairy steers (initial weight: 119 kg, s.d. 24 kg) of Swedish Holstein (30) and Swedish red breed (33) were blocked according to weight into three blocks and housed in nine pens, three pens per block, with straw bedding. In Exp. 2, 48 dairy steers (initial weight: 414 kg, s.d. 26 kg) of Swedish Holstein (22) and Swedish red breed (26) were blocked according to weight into two blocks and housed in twelve pens, four pens per block, with slatted floors. Both randomized complete block experiments lasted for twelve weeks. Initial and final weights were registered on three consecutive days. Pens within block were randomly assigned to one of three treatments: whole-crop barley silage harvested at heading, milk- or dough stage of maturity. In Exp. 1, forages were fed in a total mixed ration together with concentrate consisting of 40% rolled barley and 60% soybean meal. Feeds were mixed and fed with the goal of providing animals with 1 kg supplementary concentrate head<sup>-1</sup> d<sup>-1</sup>. In Exp. 2, forages were fed separately and supplemented with 0.6 kg soybean meal dressed on top of the silage. In both experiments, feeding took place once daily and orts remaining were collected three times a week. Data were statistically analysed with the general linear model procedure of Minitab15 (Minitab Inc., State College, PA, USA), using block and treatment as fixed factors in the model.

Table 1. Chemical composition of whole-crop barley silages used in Experiments 1 and 2. All values in g kg<sup>-1</sup> DM, unless stated otherwise. Mean (n = 3) and standard deviation (s.d.).

	Heading		Milk stage		Dough stage	
	Mean	s. d.	Mean	s. d.	Mean	s. d.
Dry matter	243	1.8	300	4.3	332	1.9
Ash	66	1.0	57	1.0	59	1.4
Crude protein	90	1.1	82	1.1	76	1.0
Sugar	5	1.5	46	6.5	10	6.2
Starch	40	16.4	24	12.7	177	14.0
NDF	547	3.6	508	27.0	431	9.9
<i>In vitro</i> OMD <sup>1</sup>	840	17.2	795	9.9	792	13.5
pH	3.8	0.01	3.7	0.04	3.8	0.08
NH <sub>3</sub> -N	14	0.7	11	1.4	13	0.2
Lactic acid	115	1.6	86	4.7	70	5.8
Acetic acid	25	1.1	15	0.7	16	0.3
Butyric acid	0.4	0.005	0.4	0.007	0.3	0.029
Ethanol	7	0.6	4	0.2	3	0.5

<sup>1</sup>*In-vitro* organic matter digestibility (g kg<sup>-1</sup>) after 96 hour incubation.

## Results and discussion

Silage dry matter (DM) intake was higher for dough stage silage than for heading- and milk-stage silages in both experiments (Table 2). Also, DM intake was higher for milk-stage silage than for heading silage in Exp. 1, which may be related to a greater negative effect of total acids on DM intake of young compared to older animals (Krizsan and Randby, 2007). The increased DM intake probably was related to the decreasing NDF concentration with advanced maturity of the silages. Feed intake has been shown to be negatively correlated with NDF content in the diet of cattle when rumen fill is limiting intake (Mertens, 1994), which presumably is the case in these experiments. A later maturity stage is, on the other hand, correlated with a lower NDF digestibility in whole-crop cereals (Nadeau, 2007) which probably explains the lower NDF intake at dough stage compared to heading stage in both experiments, and compared to milk stage in Exp. 1.

There was a tendency towards differences in live weight gain of the steers fed the different silages in Exp. 1. Feed conversion, expressed as DM intake kg<sup>-1</sup> live weight gain, differed among treatments, which indicates the potential of the feeds. Young steers in Exp. 1 fed the heading silage had the lowest feed conversion ratio and older steers in Exp. 2 fed the heading silage had lower feed conversion ratio than the steers fed the milk-stage silage. The results indicate a better nutritional quality for whole-crop barley harvested at heading than at milk stage and dough stage, reflected by the higher organic-matter digestibility of silage harvested at heading, as shown in Table 1.

Table 2. Intakes of dry matter (DMI) and NDF (NDFI), live weight gain (LWG) and feed conversion ratio (FCR) of steers fed whole-crop barley silages harvested at heading, milk- and dough stages of maturity.

	Heading	Milk stage	Dough stage	SEM	P - value
<i>Experiment 1 (n = 3)</i>					
DMI, g kg <sup>-1</sup> LW d <sup>-1</sup>	17.7 <sup>c</sup>	19.1 <sup>b</sup>	20.5 <sup>a</sup>	0.17	< 0.001
NDFI, g kg <sup>-1</sup> LW d <sup>-1</sup>	9.7 <sup>a</sup>	9.7 <sup>a</sup>	8.8 <sup>b</sup>	0.09	0.003
LWG, kg d <sup>-1</sup>	0.68	0.62	0.71	0.022	0.083
Silage FCR, kg DM kg <sup>-1</sup> LWG	3.7 <sup>b</sup>	4.4 <sup>a</sup>	4.3 <sup>a</sup>	0.11	0.020
<i>Experiment 2 (n = 4)</i>					
DMI, g kg <sup>-1</sup> LW d <sup>-1</sup>	17.1 <sup>b</sup>	17.5 <sup>b</sup>	20.2 <sup>a</sup>	0.19	< 0.001
NDFI, g kg <sup>-1</sup> LW d <sup>-1</sup>	9.4 <sup>a</sup>	8.9 <sup>b</sup>	8.7 <sup>b</sup>	0.09	0.002
LWG, kg d <sup>-1</sup>	0.87	0.68	0.86	0.062	0.109
Silage FCR, kg DM kg <sup>-1</sup> LWG	8.6 <sup>b</sup>	11.8 <sup>a</sup>	10.7 <sup>a,b</sup>	0.77	0.049

<sup>a,b,c</sup>Values with different subscripts within a row differ significantly ( $P < 0.05$ ).

## Conclusions

In order to achieve good animal performance it is concluded that whole-crop barley silage should be harvested at an early maturity stage (e.g. at heading), when digestibility is relatively high, resulting in efficient feed conversion. Harvest at a late maturity stage (e.g. at dough stage), when dry matter intake is high, also has a potential to improve animal performance, although not confirmed with an improved feed conversion in this study.

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# Nitrogen excretion and utilization in grassland dairy cows

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## Abstract

Between 1991 and 2005, nitrogen (N) excretion and utilization were studied in Friesian dairy cows offered ryegrass-red clover (*Lolium perenne* and *Trifolium pratense*) with a minimum of concentrate supplementation (2.63, sd 1.62, kg dry matter per cow per day). Daily N excretion in faeces, urine and milk was  $120 \pm 1.25$ ,  $188 \pm 2.13$  and  $101 \pm 0.74$  g, respectively. Results from analysis of variance show that 64, 46 and 21% of N intake was excreted in faeces, urine and milk, respectively. Statistically, the best relationship was observed between N intake and N in milk.

Keywords: grass, dairy cow, excretion, nitrogen

## Introduction

Milk and meat production models are usually based on intensive systems, with high quantities of nitrogen (N) and phosphorus (P) in the concentrate offered. Both these elements have a high environmental impact on the soil, water and atmosphere. Furthermore, in terms of live-stock health, a high N intake can result in ammonium accumulation in the rumen, reproductive problems (McCormick *et al.*, 1999), and milk composition modifications (Hermansen *et al.*, 1999).

On the other hand, extensive production models are based on grassland herbage, which is a cheap protein source with high rumen degradability (Salcedo, 2000) and for that reason it has low N efficiency. The aim of the present study was to determine the relationship between the N intake and N excreted in faeces, urine and milk in dairy cows offered perennial ryegrass and red clover (*Lolium perenne* and *Trifolium pratense*) with a minimum concentrate supplementation.

## Materials and methods

All experiments were conducted in metabolic housing facilities: faeces and urine (sampled by a 'Foley' vesicle probe) were sampled, weighed and analysed in a laboratory separately daily. At the same time, the quantity of offered and rejected feedstuffs was obtained. Minimum diet adaptation period for each experiment was at least seven days. For long term experiments (> 125 days), two or three cows of each group were also studied in the metabolic housing, using the same protocol. Grassland herbage offered was cut daily cut from the same grasslands. Energetic concentrate source barley or maize supplied approximately 80% of total amount of concentrate. Individual milk production was checked during the control days for each experimental period. Similarly, milk aliquot of the morning (7.30) and evening (16 p.m.) milking process were analyzed together on the Laboratorio Interprofesional Lechero de Cantabria (Spain) to determine N concentration.



Table 1. Supplementation, lactation state, milk production and days on study.

Concentrate	Forage	Milking days	Milk (kg)	Days on study
0 kg	Maize silage: 2.24 – 1.12 – 0 kg MS	40	23	90 days
2.6 kg	Maize silage: 2.24 – 1.12 – 0 kg MS	22	23.4	2 years: 180 d/year
2 kg	Grass silage: 0 kg MS	109	16.7	
4 kg	Grass silage: 6.5 kg MS	103	18.8	91 days
2.5 kg Barley-Sunflower; Barley-Soya; Barley-Fish	Maize forage dehydrated: 4 – 2 – 0 kg	38	26	237 days
5 kg Barley; Maize;				2 years:
2.5 kg Barley; Maize; 0 kg	-	22	23	285 d/year
4.5 kg Barley- Fish				
Barley-Soya; Barley-Cotton	-	11	22	125 days
0 kg	-	28	22	274 days
4.5 kg Barley-Fish;				
Barley-Soya; Barley-Cotton	-	22	26	90

## Results and discussion

Average N intake was 479 (sd 3.16) g per cow and day (Table 2) with a 20-25 milk production kg, which is in accordance with the recommendation in NRC (2001). For grassland herbage a dry matter intake of 11.8 (sd 2.3) kg/d, due probably to low height (20.5 sd 9.0 cm), high N content (3.17 sd 0.6%) and low concentrate offered (2.64 sd 1.6 kg). The highest N excretion was determined in urine ( $N_u = 37.1$  sd 6.3%), followed by faeces ( $N_f = 21.6$  sd 5.8%) and milk ( $N_m = 21.7$  sd 0.18%). On a linear model, N intake explained 49% of  $N_f$ . Calculated in that way for a N intake value inside the range of 191-757 g/d, N excretion is 0.317 g/g (Figure 1a) and 14.8 g N/kg DM. This value is much higher than the 2.6 g N/kg DM, value showed by Peyraud et al. (1995). Contrary, the parameter with the best relationship with  $N_u$  were milk urea (mg/dl), N intake (g/d) and ammonium in rumen (mg/dl;  $r_2 = 0.54$ ,  $r_2 = 0.46$  and  $r_2 = 0.42$ , respectively). However to include other variables as forage nutritive value (NDF and CP,  $r_2 = 0.39$ ), nutrients ingestion in kg/d (CP and starch,  $r_2 = 0.47$ ) or diet concentration (PNDR and NDF  $r_2 = 0.47$ ) don't improve determination coefficients.

Table 2. N intake, excretion and efficiency.

	Average	Max	Min	s.e.m	N
DM, kg/d	15.5	22.3	7.1	2.29	1045
Forage, %	83.6	100	57	10.1	1045
DMD diet, %	74.1	81	66.9	3.25	433
N Intake, g/d	479	757	191	102	1045
Faeces N, g/d	120	159	66	26	433
Urine N, g/d	188	286	138	44	433
Milk N, g/d	101	180	25	24	1034
Efficiency, %	21.7	47.8	3.9	0.62	1034

s.e.m.: standard error of the difference means

Milk N (intake efficiency N) showed a low average value with a wide range, probably due to the number of days in milk. Bruchem et al., (1991) also observed values near 20% in different experiments in Europe. The best relationship was determined between N utilization efficiency and N intake (Figure 1b) although with a low determination coefficient. The lowest and the highest  $r_2$  with N utilization efficiency were observed with lactation days ( $r_2 = 0.18$ ) and milk production variables ( $r_2 = 0.32$ ; according with Tamminga, 1992).

## Conclusions

N excretion on grassland dairy cows with a minimum concentrate intake has to be focus to a high energy source concentrate. On the other hand, a higher N amount ingested doesn't mean an increase of milk N efficiency.

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# Evaluation of a dairy sheep system in a Sardinian hill area based on natural pasture: milk production and feedstuff supplementation

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## Abstract

A dairy sheep system was studied during 2005 (Y1) and 2006 (Y2) in a hill area in Sardinia in order to evaluate forage production and quality, animal performance and feedstuff consumption during the lactation period. The system consisted of 6 ha of natural pasture grazed at a stocking rate of 4 ewes ha<sup>-1</sup> in Y1, and 6 ewes ha<sup>-1</sup> in Y2. Average herbage on offer was not significantly different between the two years but it showed a significantly different pattern between seasons. Herbage allowance was higher in spring of Y1 than in Y2 (4.8 and 1.3 kg DM head<sup>-1</sup> day<sup>-1</sup>,  $P < 0.001$ ). Herbage quality was higher in Y1 than in Y2 ( $P < 0.05$ ) with no significant differences between seasons. Average daily milk yield per head was higher in Y1 than Y2 ( $P < 0.001$ ), whereas milk production per hectare was greater at the higher stocking rate ( $P < 0.001$ ). During winter the supplementary feeds provided 52-83% of the animals' total energetic requirements, but these values decreased to 14-45% in spring.

Keywords: herbage availability, herbage quality, stocking rate

## Introduction

Mediterranean hill and mountain livestock systems are mainly based on the utilization of natural resources during the greatest part of the year. They are strongly influenced by climatic conditions which could represent a constraint, especially in winter, due to a shortage in herbage availability for grazing animals. In dairy sheep systems milk production occurs in this period and shepherds are forced to integrate grazing with supplementary feeding, with feed quantities that depend on the animals' physiological stage. Knowledge of the balance between natural pasture production and animals' requirements is essential to understanding the farms' dependence on external feedstuff. The reduction of herbage pasture in the animals' diet could result in the need for an increase in feed ration costs, and also decrease the specific characteristics of some cheeses (Morand Fehr *et al.*, 2007). The aim of the experiment was to set up a dairy sheep system in order to evaluate forage production and its quality, animal performance and feedstuff consumption during the lactation period.

## Material and methods

A dairy sheep system was studied during 2005 (Y1) and 2006 (Y2), in the hills of central-western Sardinia (39°N, 670 m a.s.l.) which has a Mediterranean climate. The system consisted of 6 ha of natural pasture rotationally grazed by 24 Sarda dairy ewes in Y1 (stocking rate: 4 head ha<sup>-1</sup>) and by 36 in Y2 (stocking rate: 6 head ha<sup>-1</sup>). The ewes were machine-milked twice a day from January to June. Meteorological data were monitored by a weather station located near the farm (SAR Sardegna, [www.sar.sardegna.it](http://www.sar.sardegna.it)). Herbage on offer (HO; 12 samples of 0.5 m<sup>2</sup> ha<sup>-1</sup>), and its chemical composition (crude protein (CP), NDF, ADF and ADL), assessed by Near Infrared Reflectance Spectroscopy (NIRS, Shenk and Westerhaus, 1994), were measured at the beginning of each grazing period. Average daily milk yield, number of grazing hours per day, and hay and concentrates offered as feed supplement were measured daily. Total energetic requirements per ewe (TER), expressed as French Feed Units (UFL; INRA, 1988), were calculated considering ewe body live weight

and its milk production. The data set was analysed by GLM to assess the effect of year, season and their interaction on the main variables of the farming system under study.

## Results and discussion

Meteorological data presented a different pattern during Y1 and Y2. No difference was found in total annual rainfall which was 676 mm in Y1 and 672 mm in Y2, mainly concentrated in autumn (48%). Y1 was characterized by a cold winter, whereas Y2 had a dry spring (9% of the Y2 total annual rainfall vs. 25% of Y1, Table 1).

Table 1. Average weather conditions in winter and spring during the first (Y1) and second (Y2) year of the trial. Seasonal means with standard errors in brackets.

		Winter		Spring		Probability level		
		Y1	Y2	Y1	Y2	Year	Season	Year × Season
T Min	°C	0.3 a (0.38)	1.7 b (0.28)	8.2 c (0.33)	8.2 c (0.43)	ns ( <i>P</i> = 0.06)	0.001	0.05
T Max	°C	7.9 a (0.45)	8.7 a (0.27)	19.2 b (0.57)	20.4 b (0.59)	0.04	0.001	ns
T Med	°C	4.1 a (0.36)	5.1 a (0.25)	13.6 b (0.44)	14.4 b (0.50)	0.04	0.001	ns
Rainfall	mm	181 b	185 b	168 b	60 a	ns	ns	ns

Means with different letters within row indicate differences between groups; ns = *P* > 0.05.

Average HO proved to be similar in the two years but was different between seasons (Table 2). HO quality proved to be higher in Y1 than Y2 without any differences throughout the year.

Table 2. Average herbage on offer (HO) and herbage allowance per head per day (HAL), chemical composition in winter and spring during first (Y1) and second (Y2) year of the trial. Means, with standard errors in brackets.

		Winter		Spring		Probability level		
		Y1	Y2	Y1	Y2	Year	Season	Year × Season
HO	t DM ha <sup>-1</sup>	0.62a (0.16)	1.10ab (0.14)	2.33b (0.21)	1.66ab (0.20)	ns	0.001	0.028
HAL	g head day <sup>-1</sup>	806a (105)	686a (4)	4800c (74)	1325b (50)	0.001	0.001	0.001
DM	g kg <sup>-1</sup>	209 (13)	252 (6)	228 (17)	246 (8)	ns	ns	ns
CP	g kg DM <sup>-1</sup>	184b (4.7)	149a (1.1)	173b (4.4)	147a (4.4)	0.001	ns	ns
NDF	g kg DM <sup>-1</sup>	470a (2.5)	575b (5.9)	479a (15.1)	545b (9.2)	0.001	ns	ns
ADF	g kg DM <sup>-1</sup>	215a (2.1)	326c (2.4)	276b (10.8)	315c (8.1)	0.001	0.030	0.003
ADL	g kg DM <sup>-1</sup>	55a (1.2)	72b (0.8)	67ab (4.3)	69ab (3.5)	0.047	ns	ns

Means with different letters within row indicate differences between groups; ns = *P* > 0.05.

Herbage allowance (HAL, kg DM head<sup>-1</sup> day<sup>-1</sup>) was strongly influenced by the season and rainfall, especially in spring when it dropped from 4.8 kg DM head day<sup>-1</sup> in Y1 to 1.3 kg DM head day<sup>-1</sup> in Y2 (Table 2). Daily Milk Yield was greater in Y1 than Y2 due to high herbage allowance and quality and to lower stocking rate. Milk production per hectare was higher in Y2 than in Y1 (Table 3). These data agree with Fois *et al.* (2000), who found that an increased stocking rate from 4 to 6 ewes per ha, in rain-fed Sardinian lowlands, resulted in a decrease in average daily milk production per ewe, and an increase in milk production per ha.

Table 3. Average daily milk yield, grazing time, concentrate and hay offered and daily milk production per ha, in winter and spring during first (Y1) and second (Y2) years of the trial. Means, with standard errors in brackets.

		Winter		Spring		Probability level	
		Y1	Y2	Y1	Y2	Year	Season Year × Season
Daily Milk Yield	g head day <sup>-1</sup>	948b (23)	842a (12)	1211c (22)	950b (19)	0.001	0.001 0.001
Grazing time	hours day <sup>-1</sup>	4a (0.3)	7b (0)	22c (0.6)	21c (0.7)	ns	0.001 0.001
Concentrate	g head day <sup>-1</sup>	421c (13)	262b (0.2)	195a (6)	248b (3)	0.001	0.001 0.001
Hay	g head day <sup>-1</sup>	735d (15)	658c (10)	74a (22)	214b (30)	ns	0.001 0.001
Milk Yield	kg ha <sup>-1</sup> day <sup>-1</sup>	3.79a (0.1)	5.05b (0.1)	4.84b (0.1)	5.70c (0.1)	0.001	0.001 0.030

Means with different letters within row indicate differences between groups; ns =  $P > 0.05$ .

During winter, due to low herbage availability, the ingested feed supplements supplied 83% and 52% of animal TER in Y1 and Y2, respectively (Figure 1). These values decreased in spring to 14% in Y1, and to 45% in Y2 (during which there was period of dry weather).

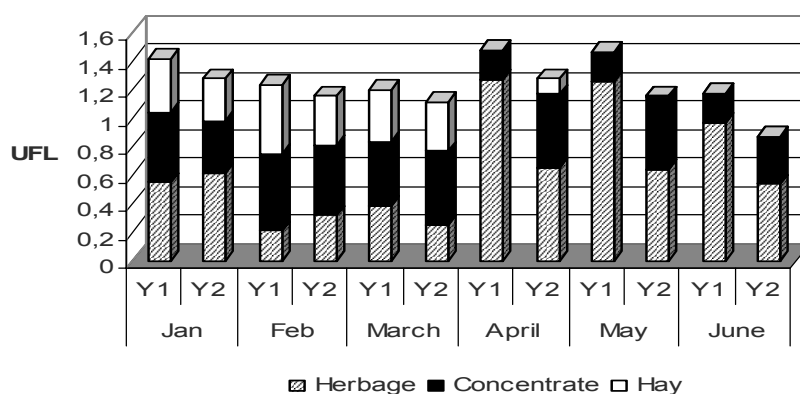


Figure 1. Pattern of the ewe's total energy requirements and energy intake from herbage, concentrate and hay measured in the first (Y1) and second year trial (Y2).

## Conclusions

The dairy sheep system in the Mediterranean hills shows an imbalance between the energetic requirements of animals and the energy available from pasture. This imbalance is most pronounced in winter when supplements could cover about 80% of ewes' TER. In spring, herbage on offer could provide up to 85% of ewes' TER depending on the rainfall. A significant rise in milk production per hectare was highlighted by increasing the stocking rate from 4 to 6 ewes per hectare.

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# Effect of mixed grazing of suckler cows and lambs on faecal egg counts and animal performance

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## Abstract

Because of high economic losses to sheep and cattle producers there is a growing need to investigate the effects of different grazing strategies on internal parasites in ruminants. This experiment was conducted over two successive years (2003 and 2004) to study the effects of grazing cultivated pasture (mainly timothy/meadow fescue/red clover) by suckler cows, either alone or together with lambs, on the faecal egg counts and animal performance. Eight Hereford suckler cow-calf pairs alone, and eight suckler cow-calf pairs together with 45 male Finnsheep lambs, grazed rotationally in two replicates from the beginning of June to the end of August. Each replication consisted of 4.2 hectares of pasture in three paddocks. The animals were not treated with anthelmintics. Faecal sampling for parasitological examinations was individually done on a monthly basis during weighing. Eggs were analysed per gram of faeces (epg) using the modified McMaster flotation method. Mixed grazing which increased the stocking rate by 21% had no negative effect on the prevalence or intensity of parasitosis. Both calves and cows grazing alone had higher live weight gains than those in the mixed grazing. However, the mixed system resulted in 120 kg ha<sup>-1</sup> higher total meat production.

Keywords: *Eimeria*, mixed grazing, parasites, pasture, sheep, suckler cow

## Introduction

Efficient and welfare-friendly livestock production requires control of helminth infections. Gastrointestinal parasites depress feed intake, cause illness and can even kill, but their main economic impact is the reduction of growth of young lambs. At present, farmers rely upon the routine use of anthelmintics to maintain health and productivity. In general, anthelmintics have proved to be highly cost-effective. However, both overuse and inappropriate use of anthelmintics has led to the emergence of parasites that are resistant to anthelmintic treatment (Jackson and Coop, 2000). Grazing management, i.e. stocking rate, rotational grazing and alternate use of pasture and livestock, has long been advocated to control nematodes without relying on effective anthelmintics. Each method has its advantages and disadvantages. If the stocking rate is low, the risk of heavy infection is limited. Rotational grazing is plausible but not very effective due to the long life of infective larvae. Alternating livestock would reduce the intensity of infection, because most species of nematodes are not able to infect different species such as sheep and cattle, i.e. they are species-specific. In spite of promising results abroad, there are no studies that have compared the effects of mixed grazing of cattle and lambs on gastrointestinal parasites within the same experiment in Nordic conditions. The main objective of this experiment was to determine the prevalence of gastrointestinal parasites in the suckler cows with their offspring alone or together with growing lambs and the effect of mixed grazing on animal performance.

## Material and methods

The experiment was carried out at MTT Agrifood Research Finland, Tohmajärvi Research Station (62°14'N, 30°21'E, 110 m above sea level) in two successive years (2003 and 2004). The timothy (*Phleum pratense* L., 65%)-meadow fescue (*Festuca pratensis* Huds., 30%) and red clover (*Trifolium pratense* L., 5%) pasture was established on sandy loam in 2001. Because the pasture was grazed for the first time in 2003, there was no residual pasture larval infection. Two grazing regimes with two replicates were used: suckler cows with their offspring alone (Treatment 1) and suckler cows with their offspring and Finnish Landrace male lambs (Treatment 2). Both replications of Treatment 1 included eight Hereford suckler cow-calf pairs, and both replications in Treatment 2 had 45 lambs. The stocking rates calculated as animal unit (AU, a 500-kg cow) were 3.2 and 3.8 AU ha<sup>-1</sup> in Treatments 1 and 2 in June 2003, respectively, and 2.9 and 3.6 AU ha<sup>-1</sup> in June 2004, respectively. Each replication was rotationally grazed on 4.2 ha from the beginning of June to the end of August for 83 days. The animals were not treated with anthelmintics. The live weights of the animals were taken at the beginning and at the end of the grazing season and at intervals of 28 days. In addition, the cows were scored for body condition according to the scoring scale from 0 to 5 twice per season. Faecal samples were individually collected directly from the rectum on a monthly basis during weighing, on average from 90 to 100% of the animals. The coccidia oocysts and helminth eggs were counted in the faecal samples per gram of faeces using the modified McMaster method with 20 eggs per gram (epg) detection level. Data on the live weight and egg counts were analysed using linear mixed models by the SAS/MIXED procedure. Animal was used as an observation unit while replication was used as an experimental unit when differences between grazing regimes were tested.

## Results and discussion

Except for July 2003, the mean daily temperature between May and September was typical of meteorological conditions recorded at this site (12.9 and 12.5 °C for 2003 and 2004, respectively) compared with a long-term (1971-2000) average of 12.1 °C. In 2003, the temperature in July was 19.5 °C and the precipitation in August twice higher than that typically recorded (160 vs. 83 mm).

At the beginning of the first grazing, 80% of the animals were infected with parasites. Coccidia oocysts (*Eimeria* spp.) were found in lambs, cows and calves (Table 1). In agreement with Gudmundsson *et al.* (1984), the numbers of coccidia oocysts in the faeces of young grazing lambs were fairly high in the early summer. Four lambs had over 50,000 coccidia oocysts per gram (opg) with a peak of 432,000 opg in June 2003. These lambs had diarrhoea. However, the number of oocysts dropped considerably during grazing and their prevalence in lambs reached minimum in August. Hot and dry weather conditions in July may have had a reducing effect on the oocyst (and nematode larval) counts on pasture and this decreased the parasitic problem. In June 2003, the mono-grazed calves were more infected with *Eimeria* oocysts than those grazing with lambs ( $P = 0.06$ ). Compared with mono-grazing, the significantly lower ( $P = 0.02$ ) *Eimeria* infection was also found in the mixed-grazing cows in August 2004. Otherwise there were no significant differences in the oocyst counts due to grazing system. The main genera of nematodes were *Strongyloides* and trichostrongylids (Trichostrongylidae family type, also called strongyle, eggs). *Strongyloides* eggs were generally present at a moderate level, except in June 2003 and 2004 when maximum counts in calves were 1700 and 2700 epg, respectively.

Before turning out in June 2003, the number of trichostrongylid (possibly *Ostertagia ostertagi*) eggs was smaller in mixed than in mono-grazed cows ( $P = 0.08$ ). The calves (max. 120 epg) and lambs (max. 40 epg) started to excrete trichostrongylid eggs in August 2003.

Because of no species identification of trichostrongylid eggs, we could not exclude the possibility that the lambs got trichostrongylid infection from the cattle, even though the possibility appears small. A statistically significant ( $P = 0.05$ ) difference in the trichostrongylid burdens in calves between treatments was recorded in June 2004.

At the end of the first grazing season, small numbers (max. 20 epg) of *Trichuris* eggs were found in cattle. Twenty percent of lambs excreted *Skrjabinema* eggs (max. 540 epg) in August 2003, but not later. *Nematodirus* eggs were discovered for the first time in cows in June 2003 and in calves and lambs in 2004. The tapeworm of sheep (*Moniezia* spp.) was not found.

Overall, the calves and cows grazing with lambs gained less weight than those grazing without lambs (Table 2). However, the difference was considered to be of no practical importance. The reduced effect on live weight gains was thought to be due to access to less pasture. According to scoring, the cows were in good condition. The growth rate of lambs was satisfactory (160 g day<sup>-1</sup>). Lamb carcasses had suitable fat scores and were adjudged to be highly acceptable. Mixed grazing with lambs increased the total meat production by 150 and 94 kg ha<sup>-1</sup> in 2003 and 2004, respectively.

Table 1. Mean numbers of *Eimeria* (coccidian) oocysts and trichostrongylid eggs per gram faeces in two successive years (2003-2004).

	<i>Eimeria</i> oocysts						Trichostrongylid eggs					
	2003			2004			2003			2004		
	June	July	Aug	June	July	Aug	June	July	Aug	June	July	Aug
Cattle alone												
Calves	623	56	138	2005	355	444	0	10	18	0	196	179
Cows	50	2	9	46	51	121	21	23	26	41	41	24
Mixed grazing												
Calves	313	64	115	2595	728	336	0	10	6	8	199	188
Cows	81	3	11	40	73	11	10	8	8	6	44	26
Lambs	21455	7880	1588	8045	4645	2377	0	0	3	0	58	72

Samples were taken on 18 June (2003) and on 8 June (2004) and at the end of July and August.

Table 2. Live weight (LW) gains and body condition scores in two successive years (2003-2004).

Year	Cattle alone		Mixed grazing		SEM <sup>1)</sup>		<i>P</i> value	
	2003	2004	2003	2004	2003	2004	2003	2004
LW gain of cows, kg/head	53	27	28	4	7.1	5.7	0.05	< 0.01
Initial body scores <sup>2)</sup> of cows	3.5	3.2	3.3	3.2	0.15	0.15	0.45	0.93
Final body scores of cows	3.5	3.3	3.4	3.0	0.15	0.16	0.77	< 0.01
LW gain of calves, kg/head	105	92	90	83	2.4	4.0	< 0.001	0.09
LW gain of lambs, kg/head			15.5	11.4				

<sup>1)</sup>SEM = standard error of means; <sup>2)</sup> Scale 0-5 (0 = thin, 5 = fat).

## Conclusions

The results showed that mixed grazing by cattle and sheep increased the stocking rate by 21%, but this was not associated with an increased parasite burden. Both calves and cows grazing alone had higher live weight gains than those grazing with lambs. However, total meat production in the mixed system was 120 kg ha<sup>-1</sup> higher.

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# What type of cow do we need for grassland based milk production?

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## Abstract

The annual milk yield per cow is not a suitable indicator for the efficiency of dairy production systems because it is strongly influenced by live weight, duration of lactation and production system. The feed conversion ratio (kg energy corrected milk (ECM) per kg dry matter intake) or the surface productivity (kg ECM ha<sup>-1</sup>) more suitable measures to judge productivity. The milk yield per cow should be expressed per unit of live weight (LW; kg ECM 100 kg<sup>-1</sup> LW) because the maintenance requirement makes up around 40% of the annual energy requirement. A dairy herd consisting of large and heavy cows requires relatively more feed surface than an equal herd of smaller cows. The comparison of large and small Swiss Brown and Swiss Fleckvieh cows in a three-year project showed a 6% higher productivity for the herd of small animals. These findings imply that there is a clear interaction between genotype and production system which should be considered in the future orientation of breeding strategies.

Keywords: dairy breeding, milk production, grass-based feeding system, feed conversion efficiency

## Introduction

A key obligation of sustainable dairy production is the conversion of roughage to milk. This is especially valid for alpine areas where natural grassland is a key resource. However, in recent years, even in natural grassland areas, dairy cattle breeding has been strongly orientated towards keeping up with the yield increase of dairy production systems largely based on corn silage and concentrates. As a consequence, farmers are strongly orientated towards high yields per cow per year. We want to show that this rather one-sided approach can be misleading and not necessarily sustainable. As in other enterprises, dairy production systems should aim at improving efficiency – the ratio between input and output. This contribution focuses on the discussion of management and cow-specific aspects and wants to show why the average milk yield is an insufficient indicator of resource use efficiency.

## Milk yield of large versus small cows

In past decades and years dairy cows have grown ever larger and heavier in the alpine region of Europe. Table 1 demonstrates how the feed conversion efficiency varies with milk yield and body weight. It is essential that the body weight is considered when the milk yield is assessed because maintenance requirements (including requirements of the foetus and changes in live weight) make up approximately 40% of the total energy requirements of dairy cows. With growing milk yield and stable live weight the maintenance requirement per kilogram of energy corrected milk (ECM) decreases, because the maintenance requirement, which is proportional to the metabolic live weight (weight<sup>0.75</sup>), can be distributed over a higher amount of milk. The highest feed conversion efficiency is therefore achieved by those cows that have the highest milk yield per unit of metabolic live weight. This is true irrespective of the production strategy (high yield strategy versus low input strategy). In a project of the Swiss College of Agriculture (SHL) the productivity of large and small dairy cows of the Swiss Brown and Swiss Fleckvieh breeds was compared for three years in a full



grazing system with seasonal calving in spring. Two herds of 9,250 kg live weight each were kept on a forage surface of 6 ha (13 heavy cows versus 16 light cows; rotational grazing with 10 paddocks each). At the start of the grazing season the small cows on average were 136 kg lighter and 10 cm smaller (withers height). With the same surface, the milk yield of the herd of small animals was higher by 6.0% in 2003 and by 6.1% in 2004 (Table 2). Arithmetically, the maintenance requirement of the herd of the large cows was about 5% lower than that of the small cows. They should therefore have achieved a higher yield. However, the herd of the large cows could not exploit this potential because the forage consumption was clearly lower than that of the herd of small cows. Therefore the feed conversion efficiency was higher for the small animals.

### **The feeding regime is important for the milk yield**

The annual milk yield is also put into perspective if we consider the influence of the production system. With confinement feeding with TMR there is no problem achieving a milk yield of 9,000 kg with cows of high genetic merit. With a full grazing regime, such average yields are not possible because the roughage intake is limited. For example, an experiment with high yielding American Holstein cows, of which one group received TMR while the other was grazed, showed clear differences in the daily feed uptake, even though the energy concentration of the ration was comparable (Kolver and Müller, 1998). While the daily feed intake was only 19 kg dry matter (DM) for the grazing cows, it was 23.4 DM for the cows fed with TMR.

Farmers in the alpine region are used to primarily assuring that individual cows can achieve maximum yields. They therefore generously allocate the grazing surface. From the point of view of surface productivity (kg milk ha<sup>-1</sup>) this is not the correct approach because the highest yield per hectare is reached with a stocking rate that does not permit the individual cow to fully cover its feed requirement because of the strong competition (MacDonald *et al.*, 2001). Because of the higher proportion of feed needed to cover the maintenance requirements the yield of the individual animal might be slightly reduced, but the larger herd can make better use of the available biomass. Thus the efficiency of the whole system is improved.

### **Interaction between genotype and feeding system**

Recent publications show that there is an interaction between the genotype and the feeding system (Buckley *et al.*, 2005; Horan *et al.*, 2005; McCarthy *et al.*, 2007). In these studies mainly Holstein-Friesian animals from North America and New Zealand were compared. The high yielding cows from North America which are adapted to a feeding regime primarily based on corn silage and concentrates proved to be unsuitable for a full grazing regime because they had a constant energy deficit which led to a loss of weight and to fertility problems (Buckley *et al.*, 2005). On the other hand, these animals could make better use of concentrate than the genotype from New Zealand. The NZ Holstein-Friesian cows are characterized by an unusually high fertility and stable metabolism, even in a no-concentrate feeding regime. Nevertheless, their yield ability is remarkable. Per unit of liveweight they produce more energy corrected milk than the American Holstein cows and therefore achieve a better efficiency.

### **Conclusions**

The annual yield per cow is an insufficient indicator for the efficiency of dairy production systems. With the indicator kilo energy corrected milk per 100 kg metabolic liveweight the relevance of body weight for the yield potential of the whole system could be better accounted for.

The ideal cow for a full grazing system is characterized by having a high grass-conversion efficiency. She achieves this because of a high grass intake potential per unit of life weight.

Table 1. Feed conversion efficiency (kg energy corrected milk per kg dry matter intake) of dairy cows depending on body weight and milk yield <sup>2</sup>. (Average NEL content (net energy lactation): 6.3 MJ NEL/kg DM).

Body weight (kg cow <sup>-1</sup> )	Annual milk yield per cow (kg ECM <sup>1</sup> )				
	5000	6000	7000	8000	9000
350	1.19	1.28	1.35	1.41	1.45
550	1.02	1.11	1.19	1.25	1.31
750	0.91	1.00	1.08	1.14	1.20

<sup>1</sup>ECM = energy corrected milk yield.

<sup>2</sup>Assumption: for body weights of 350/550/750 kg the energy requirement including pregnancy and body weight changes are: 29.44/41.37/52.17 MJ NEL cow<sup>-1</sup> day<sup>-1</sup>.

Table 2. Surface productivity (kg ECM ha<sup>-1</sup> year<sup>-1</sup>) of two different cow types under a full grazing regime in the experiment of Burgrain LU: a herd of 13 large and a herd of 16 small cows (Swiss Brown and Swiss Fleckvieh) on a surface of 6 ha for each herd.

Year	Large cows	Small cows	Relative superiority of small cows (%)
2003	11652	12346	6.0
2004	12916	13705	6.1

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# Feed intake and digestibility of whole-crop cereal silages fed to dairy heifers

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## Abstract

Whole-crop silages of oats, six-rowed barley and two-rowed barley, harvested at heading (H), early milk (EM) and early dough (ED) stages of maturity, were fed to thirty-two heifers of the Swedish Red breed in a change-over experiment. The animals were fed the silages *ad libitum* for seventeen days, of which intake was determined during the last ten days, and then 0.95 of *ad libitum* for eleven days of which *in vivo* digestibility was determined during the last five days. Differences were considered significant if  $P_{\text{Tukey's}} < 0.05$ . Organic matter intake (OMI) in kg 100 kg<sup>-1</sup> live weight was higher for ED oats than for H oats (1.73 vs. 1.37) but higher for six-rowed barley at the H than at the ED stage (1.80 vs. 1.46). Organic matter digestibility (OMD) in g kg<sup>-1</sup> decreased from H to EM stage of oats (679 to 625) and six-rowed barley (713 to 667). Neutral detergent fibre digestibility (NDFD) decreased from H to ED stage of oats (691 to 511), from H to EM stage of six-rowed barley (758 to 603) and from EM to ED stage of two-rowed barley (681 to 574). The OMD and NDFD were higher for the barley varieties than for oats.

Keywords: *Hordeum vulgare*, *Avena sativa*, silage intake, NDF, starch, forage

## Introduction

Whole-crop cereal silage (WCCS) from barley and oats offers an alternative forage to grass silage in areas where growth of maize for silage is not feasible. The feeding value of the WCCS varies among choice of cereal species and stages of maturity at harvest (Nadeau, 2007). From heading to early dough stage WCCS can be harvested using the same machines as for harvest of grass silage. At later stages of maturity the increasing losses of kernels, both during harvest and in the gastro-intestinal tract of animals, make the windrowing of the forage impractical. The aim of this study was to evaluate the effects of cereal species/varieties, maturity stage at harvest and their interaction on voluntary feed intake and digestibility in dairy heifers when fed as WCCS.

## Materials and methods

Oats (*Avena sativa* L., cv. Cilla) and six-rowed barley (*Hordeum vulgare* L., var. *vulgare*, cv. Olsok) were harvested at the heading (barley July 17, oats July 23), early milk (barley July 30, oats July 31) and early dough (barley August 7, oats August 18) stages, corresponding to maturity stages 59, 73 and 83 respectively, in the decimal code proposed by Zadoks *et al.* (1974). Two-rowed barley (*H.v.*, var. *distichum*, cv. Pasadena) was harvested at the early milk (decimal code 71; June 25) and early dough (decimal code 83; July 7) stages. All crops were harvested as big bales and Kofasil<sup>®</sup> Ultra (sodium nitrite, hexamine, sodium benzoate, sodium propionate; Hansson & Möhring, Halmstad, Sweden) was added to all the forages at an

average rate of 5 l Mg<sup>-1</sup> fresh matter. Chemical composition of the silages is presented in Table 1.

Table 1. Dry matter (DM) content (g kg<sup>-1</sup>) and chemical composition (g kg<sup>-1</sup> DM) of whole crop cereal silages of oats (O), six-rowed barley (6B) and two-rowed barley (2B) harvested at the heading (H), early milk (EM) and early dough (ED) stages of maturity.

	O H	O EM	O ED	6B H	6B EM	6B ED	2B EM	2B ED	SEM
DM	230	245	311	369	356	416	369	417	10.7
Ash	153	126	99	164	141	110	76	56	12.8
CP <sup>1</sup>	113	114	97	129	124	100	128	111	2.4
NDF	527	530	442	500	433	411	503	451	7.3
ADF	346	346	289	307	277	263	318	280	7.0
Lignin	56	60	52	44	53	51	47	48	2.2
Starch	7	16	149	2	63	162	8	140	7.7
WSC	4	8	17	33	50	33	123	85	5.6

<sup>1</sup>CP = crude protein, NDF = neutral detergent fibre, ADF = acid detergent fibre, WSC = water-soluble carbohydrates.

The animal experiment, including 32 Swedish Red heifers (326 ± 55 kg), was designed as an incomplete change-over trial, with three 4-week periods and eight dietary treatments. The heifers were fed the WCCS *ad libitum* (1.05-1.10 times the voluntary silage intake) from days 1 to 17, where intake was registered during the last ten days. The animals were fed WCCS at 0.95 times the *ad libitum* intake from days 18-28 and total faecal collection was performed from all the heifers from day 24 to 28 to determine apparent *in vivo* digestibility. In addition to WCCS all heifers were fed allowances of 0.4 kg of soybean meal day<sup>-1</sup>, 0.1 kg of minerals day<sup>-1</sup> and 0.1 kg of a vitamin premix twice a week. The statistical analyses were performed using the MIXED procedure in SAS and the statistical model used was:

$$Y_{ijklmn} = \mu + P_i + B_j + S/V_k + M_l + S/V * M_{kl} + H_{j(m)} + e_{ijklmn}$$

where  $Y_{ijklmn}$  = dependent variable;  $\mu$  = general mean;  $P_i$  = effect of  $i^{\text{th}}$  period ( $i = 1, 3$ );  $B_j$  = effect of  $j^{\text{th}}$  block ( $j = 1, 8$ );  $S/V_k$  = effect of  $k^{\text{th}}$  species/variety ( $k = 1, 3$ );  $M_l$  = effect of  $l^{\text{th}}$  maturity stage ( $l = 1, 3$ );  $S/V * M_{kl}$  = effect of interaction between  $k^{\text{th}}$  species/variety and  $l^{\text{th}}$  maturity stage ( $kl = 1, 8$ );  $H_{j(m)}$  = effect of  $m^{\text{th}}$  heifer within  $j^{\text{th}}$  block ( $m = 1, 4$ );  $e_{ijklmn}$  = residual error. Pair-wise t-tests were performed with Tukey's adjustment.

## Results and discussion

Organic matter (OM) intake varied both among maturity stages and species/varieties (Table 2). The OM intake for oats was lower at heading and early milk stage than at early dough stage despite decreasing digestibilities of both OM and neutral detergent fibre (NDF) with delayed maturity. The level of OM and NDF intake for animals eating oats harvested at the early dough stage was not exceptionally high for this kind of animal, where OM and NDF intakes of 2 and 1 kg 100 kg<sup>-1</sup> live weight, respectively, can be expected. The OM intakes of the two earlier oat harvests, however, were remarkably low. It is likely that the OM intake of oats at heading and early milk stage was limited by the low DM content in the silage (Huhtanen *et al.*, 2007). The OM and NDF intake of six-rowed barley were higher at heading than at early milk and early dough stage. This is in agreement with higher digestibilities of both OM and NDF of the silage harvested at heading. The OM intake of six-rowed barley at early milk and early dough stage was lower than expected and this could have been caused by the sharp awns on the crop (Christensen *et al.*, 1977). At heading the awns seem to be softer and may not cause the animals as much discomfort as at later harvests. The amount of awns is

less on two-rowed barley and OM and NDF intake of these silages were numerically (but not significantly) higher than for the corresponding maturity stages of six-rowed barley silage.

Table 2. *Ad libitum* whole-crop cereal silage (WCCS) intake (kg 100 kg<sup>-1</sup> live weight) and *in vivo* apparent digestibility (g kg<sup>-1</sup>) of WCCS of oats (O), six-rowed barley (6B) and two-rowed barley (2B) harvested at the heading (H), early milk (EM) and early dough (ED) stages of maturity, and average heifer live weight in kg (standard deviation in parenthesis).

Feed intake	O H	O EM	O ED	6B H	6B EM	6B ED	2B EM	2B ED	SEM
OM <sup>1</sup>	1.37 <sup>a</sup>	1.45 <sup>ab</sup>	1.73 <sup>cd</sup>	1.80 <sup>d</sup>	1.52 <sup>abc</sup>	1.46 <sup>ab</sup>	1.69 <sup>bcd</sup>	1.66 <sup>bcd</sup>	0.06
NDF	0.87 <sup>b</sup>	0.85 <sup>ab</sup>	0.87 <sup>b</sup>	1.06 <sup>c</sup>	0.78 <sup>ab</sup>	0.70 <sup>a</sup>	0.90 <sup>b</sup>	0.76 <sup>ab</sup>	0.03
Digestibility									
OM	679 <sup>c</sup>	625 <sup>a</sup>	636 <sup>ab</sup>	713 <sup>d</sup>	667 <sup>bc</sup>	675 <sup>c</sup>	693 <sup>cd</sup>	667 <sup>bc</sup>	7
Starch	nd <sup>2</sup>	970	977	nd	979	948	nd	968	9
NDF	691 <sup>c</sup>	592 <sup>b</sup>	511 <sup>a</sup>	758 <sup>d</sup>	603 <sup>b</sup>	576 <sup>b</sup>	681 <sup>c</sup>	574 <sup>b</sup>	10
Live weight	286 (32)	358 (35)	356 (76)	365 (69)	341 (33)	353 (38)	423 (48)	367 (48)	-

<sup>1</sup>OM = organic matter, NDF = neutral detergent fibre.

<sup>2</sup>nd = starch digestibility was not determined for WCCS with starch concentration lower than 10 g kg<sup>-1</sup> DM.

<sup>a-d</sup>means within the same row with different superscripts are significantly different ( $P_{\text{tukey}} < 0.05$ ).

The OM digestibility of oats was lower than that of both barley varieties at each maturity stage, though at early dough stage two-rowed barley was only numerically higher. Moreover, the NDF digestibility was lower for oats than for barley with the exception of six-rowed barley at the early milk stage. Starch digestibility was high and did not differ among species/varieties or maturity stages at harvest. The starch concentration in the crop was too low to compensate for the decrease in NDF digestibility and, as a result, the OM digestibility decreased with delayed maturity stage from heading to early milk for oats and six-rowed barley.

## Conclusions

The OM and NDF digestibilities of WCCS decrease from heading to early dough stage, whereas starch digestibility remains similar. Barley as WCCS is more digestible than oats at similar maturity stages. The difference in digestibility and chemical content between six-rowed and two-rowed barley was small, but at maturities later than heading, two-rowed barley might be more suitable as WCCS due to less amount of awns.

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# The effect of intake of total and digestible fibre on milk fat production in cows

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## Abstract

Whole-crop barley silages (WCBS) harvested at heading (BSH), milk (BSM) and dough stage (BSD) were fed in five diets to fifteen multiparous dairy cows in a change-over design over three periods of four weeks each. All diets contained 12 kg concentrate, but differed in forage as follows; Diet B1: 4 kg dry matter (DM) grass silage (GS) + *ad libitum* access to BSH; Diet B2: 4 kg DM GS + *ad libitum* access to BSM; Diet B3: 4 kg DM GS + *ad libitum* access to BSD; Diet M1: *ad libitum* access to a 0.3:0.7 mix (DM basis) of BSH:GS; Diet M2: *ad libitum* access to a 0.7:0.3 mix (DM basis) of BSH:GS. The neutral detergent fibre (NDF) intake was higher with diet B1 compared to B2 and B3. The NDF digestibility was highest for diet B1, intermediate for B2 and lowest for B3. Daily milk fat yield decreased with increasing maturity stage of the WCBS. There was no significant difference among diets in kg consumed NDF per kg produced milk fat (5.0-5.3), but kg consumed digestible NDF per kg milk fat produced was higher with diet B1 (3.8) intermediate with B2 (3.3) and lowest with B3 (3.0).

Keywords: NDF, dairy cows, whole crop cereal silage, *Hordeum vulgare*, milk production

## Introduction

The milk fat concentration in milk can be regulated with the composition of the diets fed to the dairy cows. Diets low in fibre and high in starch usually results in milk fat depression. This study examined the effect of neutral detergent fibre (NDF) intake and digestible NDF intake on milk fat yield in mid-late lactating dairy cows. The starch intake was also taken into consideration.

## Materials and methods

Six-rowed barley (*Hordeum vulgare* L., var. *vulgare*) was cut at heading (July 17), early milk (July 30) and early dough stages (August 7), corresponding to maturity stages 59, 73 and 83, respectively, in the decimal code (Zadoks *et al.*, 1974). The barley crops were ensiled as round bales and Kofasil® Ultra (Hansson & Möhring, Halmstad, Sweden) was added during baling at an average concentration of 5 l Mg<sup>-1</sup> fresh matter. The crop from a grass (*Phleum pratense* L., *Festuca pratensis* Huds.), and clover (*Trifolium pratense* L., *T. repens* L.) sward was cut, precision chopped and stored in a bunker silo. Promyr™ (Perstorp Inc., Perstorp, Sweden) was added during chopping at an average concentration of 4 l Mg<sup>-1</sup> fresh matter. The silages were fed to dairy cows in five different diets in a change-over design with three experimental periods, each four weeks long. Fifteen multiparous dairy cows of the Swedish Red breed were divided into three groups of five animals each according to their milk yields two weeks before the start of the trial.

All diets contained whole crop barley silage (WCBS), grass silage and a fixed amount of two different concentrates, A (based on wheat; 9 kg day<sup>-1</sup>) and B (based on maize; 3 kg day<sup>-1</sup>). Chemical composition of silages and concentrates are shown in Table 1. In three of the diets WCBS harvested at heading (diet B1), early milk stage (diet B2) or early dough stage (diet

B3) was fed *ad libitum* (5-10% weigh back) separately from the grass silage (fixed amount of 4 kg dry matter (DM) day<sup>-1</sup> in all three diets). In the other two diets WCBS harvested at heading and grass silage were mixed in a mixer wagon at a DM ratio of either 0.30:0.70 (diet M1) or 0.70:0.30 (diet M2). The mixes were fed *ad libitum* (5-10% weigh back). Feed intake and milk production (2 \* morning and 2 \* evening milk samples week<sup>-1</sup>) was measured during the last two weeks of each experimental period. During the last five days of each experimental period *in vivo* apparent diet digestibility was measured using acid insoluble ash (AIA) as an internal marker (Van Keulen and Young, 1977). The faecal samples were collected by voluntary defecation twice daily.

Table 1. Mean (standard deviation in parenthesis) dry matter (DM) content (g kg<sup>-1</sup>) and chemical composition (g kg<sup>-1</sup> DM) of whole crop barley silages (WCBS), grass silage and concentrates (A and B) fed in five diets to dairy cows (n = 3).

	WCBS heading	WCBS early milk	WCBS early dough	Grass silage	Conc A <sup>1</sup>	Conc B
DM	335 (21)	390 (25)	407 (12)	261 (16)	889 (5)	890 (6)
Ash	147 (15)	170 (29)	101 (18)	97 (17)	62 (2)	70 (2)
CP <sup>2</sup>	132 (2)	116 (3)	99 (2)	160 (6)	198 (2)	200 (1)
Starch	Na	71 (12)	162 (11)	na	293 (16)	300 (19)
NDF	493 (9)	407 (11)	432 (18)	449 (27)	225 (10)	180 (3)
AIA	62 (8)	98 (29)	49 (19)	27 (11)	5 (1)	4 (1)

<sup>1</sup>Concentrate A is based on wheat and concentrate B is based on maize.

<sup>2</sup>CP = crude protein, na = not analysed, NDF = neutral detergent fibre, AIA = acid insoluble ash.

All statistical analyses were performed using the MIXED procedure in SAS. Feed intake, diet digestibility and milk production were analysed according to the following statistical model:

$$Y_{ijklm} = \mu + P_i + B_j + D_k + C_{l(j)} + P*D_{ik} + e_{ijklm}$$

where  $Y_{ijklm}$  = dependent variable;  $\mu$  = general mean;  $P_i$  = fixed effect of  $i^{\text{th}}$  period ( $i = 1, 3$ );  $B_j$  = fixed effect of  $j^{\text{th}}$  block ( $j = 1, 3$ );  $D_k$  = fixed effect of  $k^{\text{th}}$  diet ( $k = 1, 5$ );  $C_{l(j)}$  = random effect of  $l^{\text{th}}$  cow within  $j^{\text{th}}$  block ( $l = 1, 5$ );  $P*D_{ik}$  = fixed effect of interaction between  $i^{\text{th}}$  period and  $k^{\text{th}}$  diet;  $e_{ijklm}$  = random residual error. Pair-wise comparisons were performed and the differences were considered significant if  $P < 0.05$ .

## Results and discussion

There were no significant differences in dietary DM intake among diets, but digestible DM intake was lower with the diet containing WCBS harvested at early dough stage (B3) as a result of lower DM digestibility of that diet (Table 2). The NDF intake and digestible NDF intake was higher for the three diets containing WCBS harvested at heading (B1, M1, M2) than for the other two diets. Starch intake increased with increasing maturity stage of the WCBS, whereas the NDF digestibility and milk fat yield decreased with increasing maturity stage. The amount of dietary NDF intake per kg milk fat yield were similar for all diets, whereas digestible NDF intake was higher for each kg milk fat produced when feeding diets B1, M1 and M2. This indicates that it is the NDF intake rather than the digestible NDF intake that affects the fat content in milk. In diet B3 the starch intake was on average 0.20 of the diet, which is the advised upper limit in dairy cow rations. However, since part of the starch was the more slowly degradable maize starch, it is possible that the high starch content in the diet did not depress milk fat content to any larger extent. Moreover, the structure of the WCBS was coarse and the particle size long, which probably had a positive effect on rumination and the buffering capacity of the rumen.



Table 2. Dietary and digestible intake (g 100 kg<sup>-1</sup> live weight), *in-vivo* apparent diet digestibility (g kg<sup>-1</sup>), milk and fat yield (kg day<sup>-1</sup>), feed efficiency and average animal live weight (kg) for cows fed five different diets.

	B1	B2	B3	M1	M2	SEM
Dietary intake						
Dry matter	3.27	3.29	3.18	3.29	3.24	0.07
NDF <sup>1</sup>	1.10 <sup>b</sup>	1.03 <sup>a</sup>	1.00 <sup>a</sup>	1.10 <sup>b</sup>	1.11 <sup>b</sup>	0.03
Starch	0.51 <sup>a</sup>	0.58 <sup>b</sup>	0.65 <sup>c</sup>	0.50 <sup>a</sup>	0.50 <sup>a</sup>	0.01
Digestible intake						
Dry matter	2.51 <sup>c</sup>	2.41 <sup>b</sup>	2.21 <sup>a</sup>	2.43 <sup>b</sup>	2.42 <sup>b</sup>	0.05
NDF	0.80 <sup>d</sup>	0.67 <sup>b</sup>	0.57 <sup>a</sup>	0.75 <sup>c</sup>	0.77 <sup>cd</sup>	0.02
Diet digestibility						
Dry matter	768 <sup>c</sup>	735 <sup>b</sup>	695 <sup>a</sup>	744 <sup>b</sup>	747 <sup>b</sup>	6
NDF	725 <sup>d</sup>	650 <sup>b</sup>	568 <sup>a</sup>	687 <sup>c</sup>	697 <sup>c</sup>	10
Yield						
Milk	27.2 <sup>bc</sup>	26.1 <sup>ab</sup>	25.9 <sup>a</sup>	26.9 <sup>abc</sup>	27.8 <sup>c</sup>	0.8
Milk fat	1.33 <sup>c</sup>	1.27 <sup>b</sup>	1.18 <sup>a</sup>	1.35 <sup>c</sup>	1.34 <sup>c</sup>	0.04
Feed efficiency						
kg NDF kg <sup>-1</sup> fat	5.2	5.0	5.3	5.2	5.2	0.2
kg DNDF kg <sup>-1</sup> fat	3.8 <sup>d</sup>	3.3 <sup>b</sup>	3.0 <sup>a</sup>	3.5 <sup>c</sup>	3.6 <sup>cd</sup>	0.1
Live weight (kg)	618	619	635	628	607	-

<sup>1</sup>NDF = neutral detergent fibre, DNDF = digestible NDF.

<sup>a-d</sup> means within a row with different letters are differ significantly ( $P < 0.05$ ).

## Conclusions

The results in this experiment suggests that it is the total intake of NDF rather than the intake of digestible NDF that affect milk fat yield in mid-late lactating dairy cows.

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# **Influence of different participation of forages from grasslands in feeding ratio on effectiveness of dairy cattle feeding**

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## **Abstract**

During the years 2003-2006 a study was conducted on three dairy cattle breeding farms (Jastrzębiec, Falenty and Biebrza) that have different systems of feeding, forage types, and levels of animal performance. The aim of study was the evaluation of utilization of forages from permanent grassland in dairy cow feeding. The demand of net energy (UFL) for the production of 1kg of milk during each of two periods (summer and winter) of feeding (Biebrza and Falenty), and in the first and second halves of lactation (Jastrzębiec), and the allocation of feed from grasslands, arable lands and concentrates, were calculated. The highest consumption of feeds per kg of milk was recorded at Jastrzębiec (indoor feeding), lower at Falenty (periodical grazing) and the lowest at Biebrza (all-day grazing). The winter system of feeding at Falenty and Biebrza was the least effective (higher UFL energy consumption per kg of milk) than the summer system of feeding. At Jastrzębiec, in both periods, in spite of high contributions of concentrates in the feed, there was significantly lower consumption of energy UFL per kg of milk produced, than for winter feeding at Biebrza and Falenty.

Keywords: cows, effectiveness of feeding, feeds from grasslands, systems of feeding

## **Introduction**

The cheapest and the most valuable fodder from grasslands is green forage from pasture (Okularczyk, 2002; Wasilewski, 2002). This fodder, with the addition of concentrates, can be the basis for summer feeding of dairy cows giving 5,500 kg of milk (Nałęcz-Torwacka *et al.*, 2001). However, for high performing cows its importance, as with hay, diminishes. Green fodder and hay are usually replaced by fodders from arable lands (maize, alfalfa) fed either as green forage or silage (Barszczewski, 2004). These changes are also accompanied by changes in the system of feeding. The aim of study was to analyse the utilization of bulky feeds from permanent grasslands for farms that are using different systems of dairy cow feeding.

## **Materials and methods**

During the years 2003-2006, a study was conducted on three different farms in Poland that specialized in dairy cattle breeding: Biebrza, Falenty and Jastrzębiec. The three farms are significantly different in terms of their area and structure of agriculture land (AL). The Biebrza farm has a large area with a high proportion (> 70% of total AL) of grasslands, and pastures are > 15% of AL. Stocking density is low with 1.42 ha of meadows and 0.39 ha of pastures per head of cattle. At the Falenty farm grasslands are 47.7% of total AL, but pastures are only 8.6% of AL, and there are 0.62 ha of meadows and 0.14 ha pastures per head of cattle. Arable lands cover 52% of AL, and are used for the production of bulky feeds and cereals. At Jastrzębiec, the proportion of grasslands is only 7.8% of AL, and there are only 0.13 ha of meadows and 1.48 ha arable land per head of cattle, and no pastures. At all farms dairy cattle breeding is carried out. Cattle are of a black-white breed with additional HF blood: *ca.* 60% at Biebrza, 70% at Falenty, and > 85% at Jastrzębiec. Mean annual milk production in 2006 (influenced both by the genetic value of herd and by feeding) was: 5,923

kg cow<sup>-1</sup> at Biebrza, 7,397 kg cow<sup>-1</sup> at Falenty, and 9,550 kg cow<sup>-1</sup> at Jastrzębiec. At the Biebrza farm (since 2005) and at Falenty (since 1999) feeds have been given as PMR, sufficient for production of about 16 kg of milk (Falenty) and about 14 kg of milk only in the winter feeding period (Biebrza). High performing cows are given additional concentrates according to their individual demand. At Jastrzębiec TMR technology is used; total feed mixtures are given separately to cows in the first and second halves of lactation, and to dry cows. On the basis of a decade of feeding experience on these farms, the composition of the feeding ratios for summer and winter feeding (Biebrza and Falenty farms) and period of feeding in the first and second half of lactation (Jastrzębiec) were determined. Feed consumption (in terms of net energy) for production of 1 kg of milk with regard to feeds from grasslands, arable lands and concentrates was calculated. The value of feeds in UFL, were derived according to INRA norms (Instytut Zootechniki Kraków, 2001).

## Results and discussion

At Biebrza pasture made an important contribution to feeding, about 70-80% of the animals' daily feeding ration. The basis of winter feeding was grass-silage (42-51% of daily ration) and maize silage (27-42%). The share of concentrates in the pasture-feeding period increased from about 15% in 2003 to 32% in 2006, while in the winter feeding it increased from 13% to 21%. At Falenty the pasture played a significantly lower part. Grazing was limited to about 4 hours per day, and the effect of this was that the share of green forage in the feed decreased from 34% in 2003 to about 16% in 2005; and in 2006, because of drought, it was only 2.2%. In the pasture season cows were given additional silage from arable land, and also grass silage plus small amounts of meadow hay. The share of concentrates in the feeding ration in 2003 was 24% in the pasture season and 23% in the winter season, and in 2006 it was 44% (summer) and 34% (winter feeding). At Jastrzębiec pasture feeding was not used at all. Grass silage, maize silage and fresh malt residues were given as roughages. The share of concentrates in the feed supplied to the cows in the first half of lactation was 43-52%, and for cows in the second half of lactation it was 29-40%.

The consumption of total feeds (expressed in terms of net energy) for the production of 1 kg of milk was different for the individual farms (Table 1). The highest, > 1 UFL, was at the Biebrza farm, particularly in winter feeding. This could be a result of incorrectly balanced feeding rations, i.e. too small a contribution of energy-rich feeds such as maize or beet pulp, and to inferior genetic quality of the herd (this herd had the lowest amount of HF blood). Feed consumption for the production of 1 kg of milk on the other two farms was from 0.66 UFL at Jastrzębiec (second half of lactation in 2005) to 0.97 UFL at Falenty (winter feeding in 2003). In all farms there was evidence of a tendency for a decrease of UFL consumption per kg of milk production. The consumption of feeds, especially roughages from grasslands, was different. The highest was at Biebrza, where 65-83% of energy used for production of 1 kg of milk in summer was from feeds from grasslands, usually green pasture forage but also 45-56% from grass silage and hay for winter feeding. At Falenty the energy consumed with feeds from grasslands, mainly from green pasture forage, was 20-35% of the daily ration in summer, and in the winter feeding period it was 10-22% of all feed. On this farm, roughages from arable lands made an important contribution. Their consumption for the production of 1 kg of milk in summer was about 40%, and in winter feeding close 60%, of the energy delivered in the daily feeding ration. At Jastrzębiec the share of roughages from grasslands was significantly lower and amounted less than 20% in the first half of lactation and 18-36% in the second period of lactation. However, the consumption of concentrates in this farm was lower. The contribution of energy from concentrates in cows feeding in first half of lactation was over 50% of energy of the total feeding ration, and in the second half of lactation it was 34-46%.

Lower consumption of energy for production of milk at Jastrzębiec and Falenty, in relation to Biebrza, depended from not only the type and quality of feeds and better balanced rations, but also on the genetic value of the herd. The cows in Biebrza, in spite of lower performance, had better health and a longer period of utilization, which confirms the advisability of using this way of feeding and utilization.

Table 1. The energy consumption in feed unit for lactation (UFL) per production of 1 kg of milk in the three analysed farms.

Year	2003	2004	2005	2006	2003	2004	2005	2006
Biebrza	SF	SF	SF	SF	WF	WF	WF	WF
All feeding ratio, in this:	0.89	0.75	0.71	0.89	1.03	0.88	1.02	0.76
- green forage	0.71	0.52	0.48	0.53	0.0	0.0	0.0	0.0
- silage + hay from PG	0.03	0.09	0.0	0.05	0.58	0.41	0.49	0.33
- bulky feeds from AL	0.0	0.0	0.01	0.0	0.28	0.35	0.27	0.28
- concentrates	0.15	0.14	0.22	0.31	0.17	0.15	0.26	0.14
Falenty	SF	SF	SF	SF	WF	WF	WF	WF
All feeding ratio, in this:	0.80	0.84	0.71	0.75	0.97	0.76	0.90	0.74
- green forage	0.28	0.18	0.12	0.02	0.0	0.0	0.0	0.0
- silage + hay from PG	0.0	0.06	0.10	0.13	0.14	0.17	0.09	0.10
- bulky feeds from AL	0.30	0.26	0.23	0.23	0.56	0.34	0.49	0.35
- concentrates	0.22	0.34	0.26	0.37	0.27	0.25	0.32	0.29
Jastrzębiec	I HL	I HL	I HL	I HL	II HL	II HL	II HL	II HL
All feeding ratio, in this:	0.89	0.77	0.81	0.80	0.77	0.74	0.66	0.71
- green forage	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
- silage + hay from PG	0.11	0.15	0.11	0.12	0.20	0.27	0.12	0.13
- bulky feeds from AL	0.27	0.24	0.30	0.30	0.24	0.22	0.25	0.25
- concentrates	0.51	0.39	0.40	0.39	0.33	0.25	0.29	0.33

SF = summer feeding; WF = winter feeding; IHL- I half of lactation; IIHL - II half of lactation.

## Conclusions

Among three studied systems of feeding in years 2003-06 the lowest energy consumption per kg of milk produced in the summer-feeding period was at Falenty, next was at Biebrza, and the highest at Jastrzębiec. In winter feeding, the lowest energy consumption was at Jastrzębiec and the highest at Biebrza. The effectiveness of different systems of feeding of dairy cows depends on not only the amount and type of fodder but also their costs. The lowest costs of production per kg of milk were at the farm in Biebrza and the highest at Jastrzębiec.

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# Effect of silage maize hybrid (dry down vs. stay green) on dairy cow performance

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## Abstract

A randomized block design experiment was conducted to evaluate the effects of two contrasting silage maize hybrids (DD: dry down vs. SG: stay green) harvested at 33% dry matter (DM) on *in situ* degradation and dairy cow performance. Thirty-eight Red-HF cows were assigned to two silage treatments and individually fed. Mixtures containing one of the maize silages, grass silage and soy bean meal (70:17:13 DM basis) were offered *ad libitum*. Additionally, each cow received 7.5 kg d<sup>-1</sup> of concentrate. Intake, milk yield and milk composition were analysed from wk 1 until wk 15 of lactation. The treatment period was from wk 5 to wk 15 of lactation. Pre-treatment data (wk 1 to 4) were used as covariates. In addition, fresh silage samples were ruminally incubated to determine the rate and extent of starch and NDF degradation. Hybrid had only little effect on *in situ* degradation of starch and NDF, and no effect on the intakes of DM, NEL, starch and NDF, and the yields of milk and milk components and body weight. It was concluded that farmers should not be concerned about dry-down or stay green characteristics of silage maize, but should just choose hybrids with the best yield prospects and feeding value.

Keywords: maize silage, stay green, dry down, milk production

## Introduction

At the same whole crop dry matter content (WCDM), stay green type maize hybrids (SG) have, 1) higher grain DM content, 2) more green leaves, and 3) lower leaf plus stem DM content than dry-down type maize hybrids (DD), suggesting a more mature cob and a less mature leaf plus stem fraction. Digestibility of maize grain is reduced with increasing DM content. In addition, increased crop maturity is associated with reduced NDF digestibility and degradation rate. Therefore, the site and extent of degradation of the grain fraction of SG varieties could be different from DD varieties. Therefore, it could be hypothesized that DD and SG maize silage are different in the site and extent of degradation, nutrient utilization and animal performance. In the literature, however, there is little information on the effect of SG and DD maize silage on *in-situ* digestibility, intake and milk performance in dairy cows. Therefore, an experiment involving a lactation trial and an *in-situ* degradation study was carried out to investigate the effects of DD and SG on *in-situ* degradation, feed intake and milk production of early lactation dairy cows.

## Materials and methods

Two contrasting dry down (DD) and stay-green (SG) hybrids were grown on two adjacent plots. The hybrids were harvested at a target WCDM of 320 g kg<sup>-1</sup> (chop size 6 mm with kernel processing 2 mm) and stored in silage clamps. A lactation trial utilizing a complete randomized block design with two treatments was conducted with 38 red-HF cows. The cows were paired on the basis of similarity in parity, calving date, milk yield and milk composition during the previous lactation. Within pairs, the cows were randomly allocated to one of two



treatments: DD and SG. The experimental period was from week 1 to week 15 of lactation and consisted of a 4-week pre-treatment period followed by an 11-week treatment period. The cows were fed *ad libitum* a mixture containing maize silage, grass silage and soy bean meal (70:17:13 DM basis). Additionally, each cow received 7.5 kg DM d<sup>-1</sup> of concentrate. During the pre-treatment period, the maize silage in the feed mixture consisted of both DD and SG (50:50 DM basis). During the treatment period, treatment group DD and SG received only DD or SG maize silage, respectively. The *in situ* nylon bag degradation study was performed at the Nutreco RRC (Boxmeer, the Netherlands). For each of the maize silages, fifty nylon bags were ruminally incubated in three rumen-fistulated cows according to the protocol of the CVB (2003). The incubated maize silage samples were unground, except for particles with a size larger than 1 cm which were cut manually. After incubation, the nylon bags were pooled by incubation time and analysed for starch and NDF content. The data were fitted on the model of Ørskov and McDonald (1979):  $Y(t) = S + D(1 - e^{-k_d t}) + U$ , in which  $Y(t)$  is residue at time  $t$ ,  $S$  is the soluble fraction (%),  $D$  is the non-soluble potential degradable fraction (%),  $U$  is the undegradable fraction,  $k_d$  is the degradation rate (% h<sup>-1</sup>). The proportion of rumen-pass starch ( $B$ ) was calculated as  $B = (100 * k_p(k_p + k_d) - D) - 0.1S$  in which  $k_p$  is the rumen passage rate (% h<sup>-1</sup>). It was assumed that 10% of the soluble starch is by-pass starch (CVB, 2003). Dry matter content, total DM yields and the proportions of leaf plus stem and cobs of hybrid DD and SG were measured at harvest. Representative samples of all feeds were taken weekly for analysis on DM, CP, OM, starch, NDF, ADF and *in vitro* digestibilities of OM (dOM) and NDF (dNDF). Feeding value NEL, available protein and degradable protein balance (DVE/OEB system; Tamminga *et al.*, 1994) were calculated according to CVB (2005). Milk yield and body weight of each cow were recorded twice daily. Milk composition of each cow was determined weekly, at four consecutive milkings. Intake of nutrients, NEL, DVE, OEB were calculated from the intake and composition of feeds. The data on DMI, intake of nutrients, milk performance, and body weight were analysed using the ANOVA procedure of Genstat. Data from the pre-treatment period were used as covariates. The treatment means were compared using Student's t-test ( $P < 0.05$ ). The following model was used:  $Y_{ijk} = \mu + Y_{COV} + \text{Block}_i + \text{Variety}_j + \epsilon_{ijk}$  in which  $Y_{ijk}$  is treatment mean,  $\mu$  is total mean,  $Y_{COV}$  = covariate treatment mean pre-treatment period,  $\text{Block}_i$  is the effect of blocks ( $i = 1 \dots 19$ ),  $\text{Variety}_j$  is the effect of maize variety effect ( $j = \text{dry-down, stay-green}$ ) and  $\epsilon_{ijk}$  is the random error.

## Results and discussion

The DM content of the fresh cob fraction was higher in SG than in DD (500 vs. 545 g kg<sup>-1</sup>). The *in-vitro* dNDF was slightly lower in DD than in SG (46.3% vs. 48%). The chemical composition, *in-vitro* dOM and *in-situ* degradation characteristics of DD and SG maize silages were very similar (Table 1), which agrees with observations on SG and DD in laboratory silos (van Schooten, 2005). The  $k_d$  of *in-situ* degradation and D-fraction of starch were lower in DD than in SG, but the proportions of by-pass starch were similar. The U-fraction of NDF was smaller and the D fraction of NDF was larger in DD than in SG. Further,  $k_d$  of *in-situ* of NDF degradation was higher in DD than in SG. There were no lag-phases for the degradation of DM, starch and NDF detected. Contrary to many other studies, incubation was done with unground fresh silage samples. This explains lower rates of starch and NDF degradation than often reported in the literature. The treatment means of dairy cow performance are given in Table 2. Intake of DM and nutrients were not significantly different between DD and SG, except for OEB. The OEB was little, but significant higher for SG than for DD. However, this difference is not relevant, because in both treatments OEB was above zero, indicating sufficient nitrogen available for microbial protein synthesis. Feeding DD or SG maize silage did not affect the yield of milk, milk constituent yield and milk composition,

body weight. This is logically the result of a very similar intake and nutrient to the cows on DD and SG maize silage.

Table 1. Chemical composition and *in-situ* degradation characteristics.

	DD	SG
DM (g kg <sup>-1</sup> )	324	319
dOM (%)	75.5	74.3
Starch (g kg DM <sup>-1</sup> )	355	360
NDF (g kg DM <sup>-1</sup> )	371	371
NEL (MJ kg DM <sup>-1</sup> )	209	211
DVE (g kg DM <sup>-1</sup> )	47	45
OEB (g kg DM <sup>-1</sup> )	-36	-31
<i>in-situ</i> degradation starch		
U (%)	0.3	0.9
D (%)	30.6	36.3
S (%)	69.1	62.8
kd (% h <sup>-1</sup> )	4.0	5.4
B (%)	23.3	23.2
<i>in-situ</i> degradation NDF		
U (%)	30.8	35.4
D (%)	60.7	55.4
S (%)	8.6	9.2
kd (% h <sup>-1</sup> )	0.9	0.7

Table 2 Dairy cow performance.

	DD	SG	lsd	P
Intake				
Voluntary DMI (kg d <sup>-1</sup> )	16.0	16.0	0.8	0.95
Total DMI (kg d <sup>-1</sup> )	22.9	22.9	0.9	0.87
NEL MJ (g d <sup>-1</sup> )	157	155	0.6	0.44
DVE (g d <sup>-1</sup> )	2169	2116	86	0.21
OEB (g d <sup>-1</sup> )	137	172	16	< 0.001
NDF (kg d <sup>-1</sup> )	8.36	8.32	0.33	0.78
Starch (kg d <sup>-1</sup> )	4.46	4.54	0.20	0.40
Milk production				
Milk (kg d <sup>-1</sup> )	36.4	36.0	1.6	0.57
Fat (g d <sup>-1</sup> )	1672	1680	101	0.87
Protein (g d <sup>-1</sup> )	1243	1257	60	0.64
Body weight (kg)	638	643	11	0.91

## Conclusions

Farmers should not be concerned about dry-down or stay green characteristics of silage maize, but just choose hybrids with the best yield prospects and feeding value.

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## **Session 5**

### **The grassland landscape as a base for animal production – present, past and future**



# The historical legacy on grassland plant diversity

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## Abstract

We cannot protect species richness in semi-natural grasslands for the future without considering the temporal and spatial context in which they have evolved. During the last 100-150 years many types of grassland have been destroyed through cultivation, fertilization, abandonment or afforestation. Besides habitat destruction, fragmentation is considered to be a serious threat to species diversity in the remaining grasslands. Old maps from 200-300 years ago and more recent aerial photographs are important tools to understand grassland development in different types of landscapes. However, grassland change and rate of decline depends on the geographical properties of the landscape. Studies from different landscapes with a focus on fragmentation effects on plant species richness were compared and their divergent results discussed. Present-day species richness was related to past landscape patterns in landscapes with more than 10% grassland left, whereas present-day landscape patterns explained species richness in more fragmented landscapes. Small remnant habitats such as road verges and midfield islets are also important for plant species richness, particularly in landscapes with little semi-natural grassland left. Although past landscape patterns do not directly influence species richness today in highly transformed landscapes, it is still important to use historical geographical data to understand the fragmentation process and to devise appropriate conservation measures.

## Introduction

Semi-natural grasslands have the highest small-scale plant species richness in the world, and are key-habitats for many other organisms in agricultural landscapes. Human activities during millennia have had a strong impact on landscape patterns in Europe, and consequently much of the vegetation composition that can be seen today is the result of long-term interactions between species requirements, environmental gradients and anthropogenic management. Today, semi-natural grasslands are becoming increasingly rare and are as a consequence an issue for many conservation and restoration measures. In this paper I will first touch upon the origin of European grasslands and discuss some of the threats and conservation issues in a landscape context. Grassland diversity is threatened not only by land use change or fragmentation, but also from acidification, eutrophication, and climate change, although these threats are outside my scope here. Old maps and aerial photographs are important geographical tools to understand grassland decline, fragmentation processes and to identify other important habitats for grassland communities. However, results and the relationship between present-day plant diversity and landscape patterns need to be discussed within a spatial and temporal landscape context. Finally, I argue that grassland conservation and restoration need a broader landscape ecological perspective rather than traditional spatial ecology or conservation. My focus is on plants, although semi-natural grasslands are important for many other organisms too. Furthermore, my focus is on north-western Europe and particularly on landscapes with little semi-natural grassland left. The situation in Central and Eastern Europe is different, with more than 12% semi-natural grasslands left in the agricultural areas, compared to less than 6% in the North West (U. Emanuelsson pers com). But on the other hand, when the remaining traditional rural landscapes of Europe change, the threats to

grasslands and biodiversity will be the same, and therefore the conclusions and challenges presented here are important for grasslands and rural landscapes all over Europe.

## **How did we get here?**

### *The early history of grassland*

Many of the plant species that we today associate with managed grassland habitats such as pastures and meadows, probably existed in the pre-agricultural landscape already (Eriksson *et al.*, 2002). The European grasslands have a long history, as a more or less semi-open grazed park-like landscape (Vera, 2000) and with local naturally open grasslands on dry infertile soils and on floodplains (Svenning, 2002), and windswept coasts. Fires and shorelines during the isostatic postglacial uplift would naturally have created open grassland areas in northern Europe. These grasslands were kept open by large herbivores such as the wild antecedents of domesticated cattle and horses, bison, deer and wild boar. Human hunters probably 'managed' the grasslands by fire to improve grazing for their prey, in the same way as past hunters did on the African savannas or in Australia. Later, between 10,000-5,000 years ago, human agricultural practices started to shape the landscape more and more, and grasslands became the key habitat for the survival of the then domesticated livestock.

From the time of the Late Iron Age there was a stronger development of permanent farms and villages than in earlier times. The village or farm was surrounded by an enclosed infield area keeping the livestock away from crops and meadows. Outside the infield system there were areas of extensively grazed communal land. Hay could be transported from mires or shore-meadows far away from the village. Within the infield system all small grassland habitats were cut to provide fodder, and livestock grazed the fields after harvesting and during fallow years. During this period grasslands increased in area and connectivity, and became spatially more permanent. The spatial distribution of different parcels within the infield system was fairly stable for a long time. Swedish old cadastral maps from the 17th and 18th centuries depict a spatial grassland distribution that was established almost 1000 years earlier (Sporrong, 1971; Widgren 1983). Moreover, forest as we know it today as a closed canopy forest, is a land use that has been in practice for only 200 years. Vera (2000) argues that the terms for 'forest' in European languages in the Middle Ages did not have the same meaning we associate with the term today, but that 'forest' relates to the uncultivated wilderness that had a wide range of uses. An example of the importance of grasslands is how they were valued; in 1250 meadows in England were valued twice as high as private pasture, and four times more than arable fields (Rackham, 1994). Fodder was grown not only for local consumption but also for cash. Today we value landscapes with the focus on crop or forest production, but for thousands of years grassland was the fundamental habitat for both farmers and hunters.

Grasslands with a long continuity of management may contain an amazingly high plant species richness at small spatial scales, where it is possible to find 40-60 different plant species per m<sup>2</sup> (Kull and Zobel, 1991; Pärtel *et al.*, 1996; Eriksson and Eriksson, 1997; Austrheim *et al.*, 1999). Often small areas, a few square meters, may harbour the majority of species occurring within a semi-natural grassland site covering several hectares (Öster *et al.*, 2007). It is not clear whether it is the long continuity of grassland management *per se* or indirect effects, such as low nutrient levels, that promote the high species diversity (Eriksson *et al.*, 2002). Dispersal of plants to and between grasslands was aided by wandering livestock and transportation of hay (Poschlod and Bonn, 1998) and seeds were actively dispersed by throwing out barn sweepings to improve grasslands (Edwards *et al.*, 2007). Thus, since the Late Iron Age and possible even earlier, plant species have dispersed within and between

grasslands under the influence of grassland management, and over time an exceptional species-richness has developed.

#### *The last 100-150 years*

During the last 100-150 years the agricultural landscape has undergone dramatic changes compared with the previous 1000 years. Mechanization and draining the land made it possible to expand crop-fields on to former meadows and pastures. First, meadows and pastures on deep, usually clay-textured, soils disappeared. Fodder for the livestock was grown on fields. Forest, timber and fibre became increasingly more valuable. Pastures on thin soils and grazed semi-open forests were later abandoned or turned into forest. Small habitats, such as ponds, midfield islets, stone walls, ditches and paths have been removed (Figure 1). The variation of grassland management and intensity in the traditional landscape has changed dramatically (Gustavsson *et al.*, 2007). During the whole period industrialization and urbanization increased, although more rapidly after the Second World War. For example, 89% of the population in Sweden lived in the rural countryside 100 years ago, whereas only 15% live there today. A general pattern of intensification or abandonment of agricultural landscapes can be seen more or less all across Europe. Thousands of small farms were abandoned or incorporated into larger units. It is estimated that 90% of the semi-natural grasslands in north-western Europe have disappeared during this period (Vera, 2000; Stoate *et al.*, 2001). In Sweden there were at least 2 million hectares of semi-natural grasslands at the beginning of the 20th century, whereas there are c. 270,000 hectares left today (Persson, 2005). In the modern European landscape, semi-natural grassland fragments are left in a matrix consisting mostly of forests, agricultural fields and urban areas, although there are areas of extensive grasslands in mountain regions. This decline in grasslands is not only threatening plants but other organisms such as insects, birds, fungi and soil organisms (e.g. Rydin *et al.*, 1997; Wardle *et al.*, 1999; Weibull *et al.*, 2000; Söderström *et al.*, 2001; Stoate *et al.*, 2001).

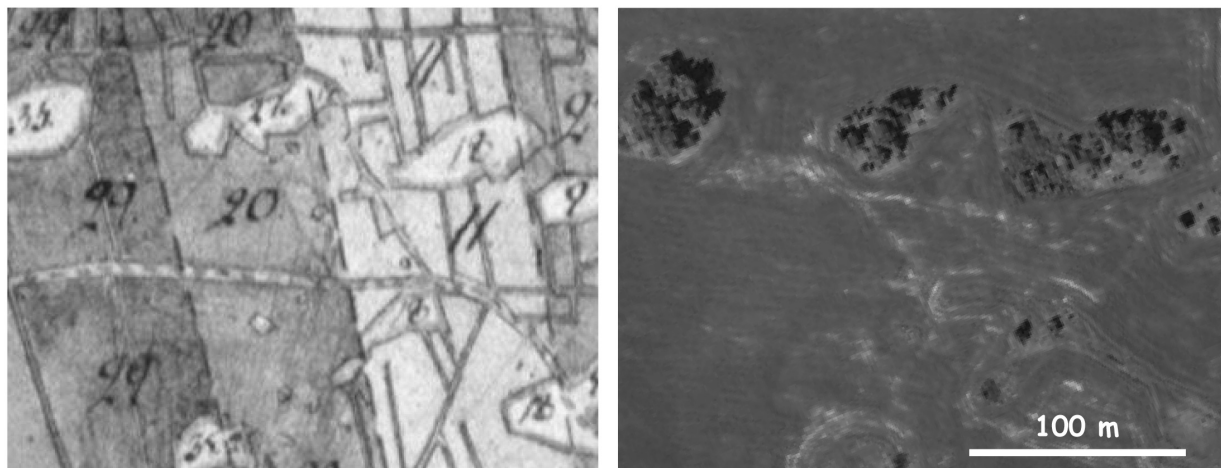


Figure 1. Old cadastral map from 1784 (left) showing crop-fields and meadows belonging to different farmers in the village, small managed grassland habitats on bedrock outcrops, ditches and a path leading to the outland (the dotted line running across the map). Each number corresponds to written records on yield and management. The right hand photo shows the same area today, with one homogenous crop-field. Only the larger outcrops remain as midfield islets, but although these have not been managed for many decades they contain a high small-scale richness with many grassland plant specialists left.



# *Grassland change in an agricultural landscape of south-eastern Sweden – a case study*

The development of grassland distribution in a landscape in south-eastern Sweden can be illustrated with layers of historical maps and aerial photographs (Figure 2). This is an area where historical records and ancient remains confirm that most of the larger farms or small villages were established more than 1000 years ago. A map from 1901 (Figure 2a) shows the distribution of arable fields, farms and cottages, and the different managed grasslands. Many changes had occurred already: primarily meadows and wetter pasture that had been drained and cultivated. A small inset map (Figure 2c) shows a cadastral map from 1688 over one village. There are old cadastral maps from most of the larger villages and farms. The remaining semi-natural grasslands today are superimposed on an orthophoto (Figure 2b).

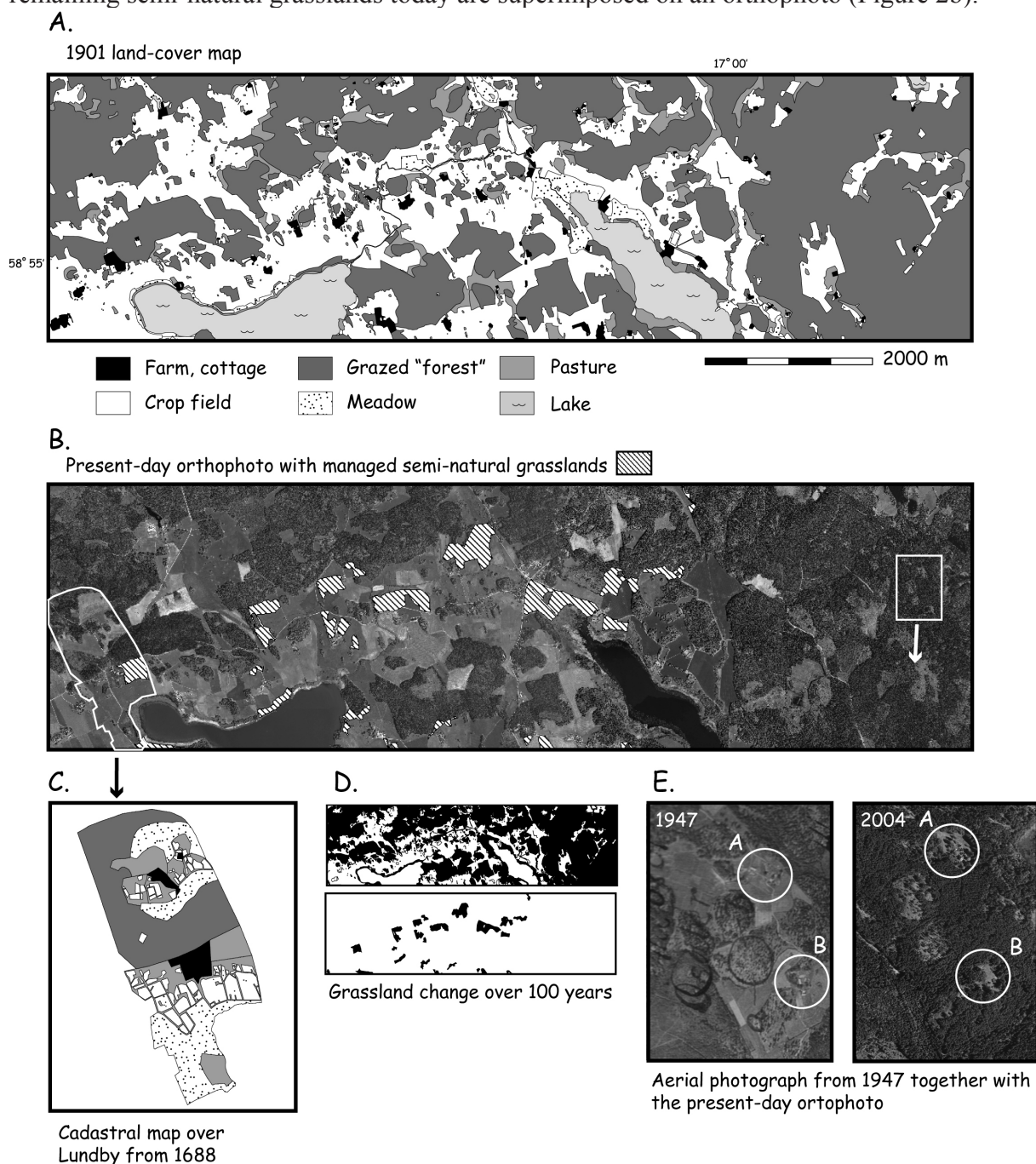


Figure 2. Grassland and land use changes in an ordinary agricultural landscape in south-eastern Sweden. See the text for detailed description. The legend beneath map (a) also applies to the old cadastral map (e).

These grasslands can be found in 17th century maps and had been managed since the Iron Age. There has been a decline in the proportion of the land area consisting of semi-natural grassland, from 60% in 1901 to less than 3% today (Figure 2d). After the first period of grassland change more than 100 years ago, there was a second period when forest grazing was banned. Forest became dense after the 1940s and farms in marginal areas were abandoned due to political decisions that all farmers would have the same income as an industrial worker (Figure 2e compared with 2b). Figure 2e shows two small crofts (A and B) with surrounding fields and grasslands in 1947, which were abandoned and pastures and fields were planted with forest. It is still possible to see openings in the dense coniferous forest with remnants of grassland vegetation, old fruit trees and garden shrubs

### **Where are we going?**

The historical legacy of past grassland management in present-day landscapes can be seen in both managed and abandoned habitats, such as the high level of small-scale species-richness in managed semi-natural grasslands. Furthermore, studies have found that plant species-richness in grassland patches today is strongly related to the historical landscape context, and in particular to grassland extent and connectivity to other semi-natural grasslands back in time (Bruun *et al.*, 2001; Lindborg and Eriksson, 2004; Helm *et al.*, 2006). The explanation behind these patterns is that many plant species show a slow response to habitat fragmentation and degradation (Eriksson, 1996; Eriksson and Ehrlén, 2001). As a result contemporary grasslands have an extinction debt (Tilman *et al.*, 1994), and particularly long-lived plants can form remnant populations that may survive for decades after environmental change (Eriksson, 1996; Dahlström *et al.*, 2006; Helm *et al.*, 2006; Lindborg, 2007). On the other hand, other studies have not found any effects of past landscape patterns on present plant-species patterns in grasslands (Bruun, 2000; Adriaens *et al.*, 2006; Cousins *et al.*, 2007; Öster *et al.*, 2007). In these studies the contemporary habitat size has been able to explain plant-species diversity.

The historical legacy can also be found in many small habitats, like forest edges, midfield islets, ditches and stone walls that have fragments of grassland communities left, although they are not managed (Tikka *et al.*, 2000; Pywell *et al.*, 2004; Cousins, 2006; Jantunen *et al.*, 2007; Cousins and Lindborg, 2008), as well as in deciduous forest (Cousins and Eriksson, 2002; Dahlström *et al.*, 2006; Ruprecht, 2006). The loss of species after abandonment occurs at a different pace for different habitats. On thin soils the encroachment of trees and shrubs is not as rapid, and therefore remnant grassland communities can persist for decades (Cousins, 2006), although many short-lived species decline relatively rapidly. Another important grassland habitat today is road-verges; the importance of this habitat for both plants and insects has been acknowledged in several countries in Europe.

The extinction debt of plants has implications for conservation of grassland habitats. If the habitat or landscape pattern remains at the *status quo*, species will become extinct in the future and species-richness will become in equilibrium with the habitat and landscape configuration. To avoid future extinctions it is necessary to apply sufficient conservation measures to restore past landscape or habitat distribution.

### **What can we do about it?**

There are extensive reviews on fragmentation effects and species diversity (Harrison and Bruna, 1999; Debinski and Holt, 2000; Fahrig, 2003; Ewers and Didham, 2006), although they mainly concern animal studies. One of the more surprising findings of an analysis of studies on fragmentation effects on plant diversity in grasslands (Cousins *ms*) was that so few studies had actually stated the time and extent of habitat fragmentation: only seven out of 61 (ISI Web of Science). Fragmentation is a process, but despite this most papers just stated that



their target habitat was 'fragmented' without providing any data on past or present habitat extent. The seven studies that had stated the habitat decline were from Belgium, Estonia and Sweden with a time-span between 70 and > 200 years (Lindborg and Eriksson, 2004; Adriaens *et al.*, 2006; Cousins, 2006; Helm *et al.*, 2006; Honnay *et al.*, 2006; Cousins *et al.*, 2007; Honnay *et al.*, 2007). Based on these studies present-day species-richness was related to past landscape or habitat patterns in landscapes with a fair amount of target habitat left; whereas in highly transformed landscapes with little grassland left, plant species richness was more related to the contemporary landscape or habitat pattern. It is possible that there is a spatial extinction threshold when grassland habitats reach below 10% or a lag effect in time (Cousins *ms*).

Most grassland studies have been conducted where there is a fair amount of grassland left or in landscapes otherwise identified as having high biological and cultural values. But to be able to find possible extinction thresholds it is necessary to conduct studies in a range of different landscapes (Cousins, 2006; Cousins *et al.*, 2007; Öster *et al.*, 2007). Landscape change trajectories might be very different depending on landscape properties, for example climate, bedrock and soil types, distance to towns, geographical location etc (Figure 3).

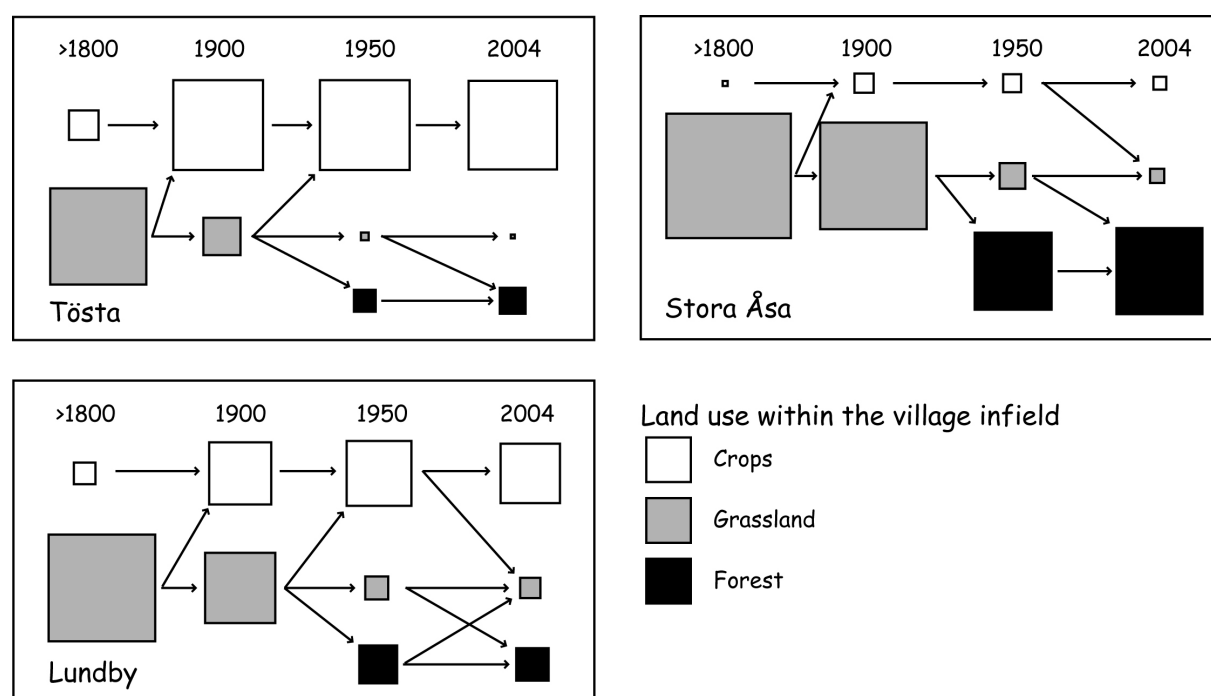


Figure 3. Old maps and aerial photographs were used to create four different time-steps in three typical Swedish landscapes. Box-size represents the percentage of land use within the village infield area. More than 200 years ago, grassland encompassed over 60% of the infield area. Most grassland disappeared before 1900 in Tösta, primarily vast areas of meadow, and today Tösta is an open modern agricultural landscape with open crop-fields and few small remnants of grasslands. Stora Åsa is situated in a region with less suitable soils for agriculture. The major grassland decline was when forest grazing was banned. In Lundby (see also Figure 1) many grassland changed into crop-fields prior to 1900 but poorer types of grassland remained until 1940s when they became forest. Today, Lundby has a fair amount of grassland as some ex-arable fields are grazed and forested areas have been restored back to grassland but there is little continuously managed semi-natural grassland left. Stora Åsa is the landscape the highest and most exclusive plant diversity today.

Recently there has been an increasing awareness within biological conservation of trying to look at larger scales, i.e. the landscape scale, when analysing status and threats to species diversity. Many studies in conservation biology rarely regard the landscape, land use or matrix surrounding the target habitats, and even more seldom the past land use or landscape pattern. There has been a tendency, and still is, among many ecologists and conservationists, to regard target or protected habitats as black blobs on an otherwise white map. However, they are not isolated islands in a hostile or inhabitable sea, but rather they are situated in a heterogeneous setting, where the matrix has different shades of grey. The main difference between landscape ecology and traditional spatial ecology is that landscape ecology not only studies what a target habitat is, but also where it is situated in both space and time. The 'where' is what places the target habitat in relation to other habitats, i.e. in a landscape context. On the other hand, it is apparent when reviewing landscape ecological literature that very few of the landscape studies also include and analyse detailed species data. To study effects of landscape pattern on species diversity is time-consuming, and to be able to compare across landscapes even more so. Therefore, meta-analysis based on results from different regions and countries will be a valuable tool to understand the effects of fragmentation. To understand the threats to grasslands in a landscape context demands a broad interdisciplinary approach, as there is a linkage between human geography, spatial statistics and analysis, remote sensing and modelling, physical geography, economy, agricultural history and, of course, ecology. There is a challenge to bridge the gap between these different disciplines.

As illustrated by Figures 1 and 2, old maps and aerial photographs have recently become more commonly used to document relationships between past and present grassland patterns (Pärtel *et al.*, 1999; Norderhaug *et al.*, 2000; Cousins and Eriksson, 2002; Lindborg and Eriksson, 2004; Gustavsson *et al.*, 2007; Öster *et al.*, 2007), and other types of fragmented habitats (Piessens and Hermy, 2006; Vellend *et al.*, 2006). Sweden and areas situated behind the Swedish borders in the 17th and 18th centuries have unique collections of old cadastral maps showing grassland patterns within the village infield system. They usually have both high thematic and spatial resolution (Figure 1, Figure 2c). Old cadastral maps have been used in several studies in Sweden to analyse past land use patterns (e.g. Skånes and Bunce, 1997; Cousins, 2001; Dahlström *et al.*, 2006; Gustavsson *et al.*, 2007), and also in other European countries, including Estonia (Pärtel *et al.*, 1999), Norway (Domaas, 2007), Switzerland (Kienast, 1993), Finland (Vuorela *et al.*, 2002; Käyhkö and Skånes, 2006), Germany (Bender *et al.*, 2005), Belgium (Petit and Lambin, 2002) and France (Dutoit *et al.*, 2004). They have also been used outside Europe, e.g., in Israel (Levin, 2006), the US (Foster and Motzkin, 2003), Japan (Ichikawa *et al.*, 2006) and Australia (Spooner and Lunt, 2004). Although even more countries have aerial photographs from the 1940s, it is still fairly unusual that this type of information is used for analysing landscape change. There are problems of generality when using old map data, i.e. classifications; the detail and accuracy might vary between landscapes and countries, and they are sometimes difficult to obtain and do not cover all areas in a region. Also interpretations and classification of aerial photographs may be quite different between interpreters, even from the same country. Still the potential for using old maps and aerial photographs is immense, particularly for making trajectories on grasslands and remnant habitats within the infield systems. It is possible for researchers from many different disciplines to meet in cross-disciplinary landscape studies.

### *Conservation and restoration*

Today much grazing occurs on former crop fields with low species-richness. Abiotic conditions on grazed former-crop fields are likely to be unsuitable for many grassland plant species due to high residual soil fertility and compaction caused by intensive agricultural practices (Pykälä, 2000; Pywell *et al.*, 2002). Still, there is spontaneous dispersal of grassland species

into grazed arable fields from small remnant habitats within the fields or from neighbouring surrounding habitats (Ruprecht, 2006; Cousins and Aggemyr, 2008; Cousins and Lindborg, 2008). It is possible that grasslands in landscapes with a more heterogeneous matrix might be more resilient to fragmentation effects than grasslands in homogenous landscapes, as small habitats such as road verges, midfield islets, or abandoned grasslands harbour remnant grassland communities. Consequently, small remnant habitats can be utilized and incorporated into larger grassland complexes to improve diversity in agricultural landscapes with few species-rich habitats left.

For the conservation of remaining semi-natural grasslands, it is important not only to understand the past extent but also the dynamics of past landscapes. The homogenization of land uses in grasslands is one factor, but the number of people who lived and utilized the landscape in the past compared to today, is another aspect that is often overlooked. People and animals contribute to plant dispersal (Peterken and Game, 1984; Kiviniemi and Eriksson, 1999; Bruun and Fritzboeger, 2002; Löfgren and Jerling, 2002; Couvreur *et al.*, 2004), and therefore connectivity is not just a matter of geographical distance between target habitats but the functional connectivity; i.e. the degree to which the landscape and managers (the matrix) facilitates or impedes movement among resource patches. Livestock moving between faraway grasslands may resume a functional connectivity, whereas grasslands situated fairly close to each other might have a low connectivity if livestock are never allowed to move in between them. If this part is overlooked it is possible that conservation measures will not be enough to preserve plant diversity for the future, even without considering the additional effects of possible extinction debts.

## **Conclusions and challenges**

The extensive historical grasslands are today occupied by forest or crop fields, with only small remnants of managed or unmanaged habitats left. Few grazed grasslands today are authentic semi-natural grasslands with a long continuity of management, as most are fertilized pastures or crop-fields. The long period of historical grassland management, without artificial fertilizers, is a prerequisite for the high species-richness and beautiful habitats that we can find in the landscape today. Semi-natural grasslands are both a natural and cultural heritage. We need to raise public awareness on the authenticity and threats to semi-natural grasslands today. Many people would probably appreciate the costs of conserving something they also value as being both beautiful or a part of our common history, and not only because of their high biological values. To understand fully the effects of fragmentation on grassland species diversity it is necessary to conduct cross-disciplinary research in all types of agricultural landscapes, and not only in landscapes with many 'hot-spots' left. The challenge for the future clearly lies with an integration of conservation at the level of species, communities and processes, and incorporating human dimensions at a landscape scale. Just how this integration should be achieved is still a problematic issue.

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# Low-intensity livestock systems in Europe: an opportunity for quality products, recreation revenues and environmental conservation

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## Abstract

Low-intensity livestock systems are important as reservoirs of biodiversity and providers of ecosystem services for society, as well as a source of income in many marginal agricultural areas. Although in most areas dairy systems continue to intensify, sheep/beef production systems tend to be low-intensity, reflecting poorer land quality or harsher climate. Low input systems can offer health and ecosystem service benefits above and beyond their intensive counterparts; given the high performance in these areas, low-input systems should not be relegated only to European marginal areas. Yet, key challenges remain, of which the most prominent may be land use change, climate changes, and long term economic viability and ecological sustainability. Current scenarios for global change conditions project significant biodiversity losses and a degradation of ecosystem services worldwide. However, the scenarios are limited in their capability to propose much-needed innovative measures to face the new situation. For both intensive and extensive systems, it is urgent to design innovative measures to face the new situation, within which low-intensity livestock systems are expected to have an increasingly important role. We explore these issues in relation to biodiversity and its role as provider of ecosystem services such as productivity, forage quality, and soil carbon storage.

Keywords: biodiversity, climate change, ecosystem goods and services, grassland management, livestock type

## Definition and origin of low-intensity livestock systems in Europe

Low-intensity agricultural systems can be defined as those systems that are used for production but managed with low levels of external inputs (fertilizers, crop protection and concentrates). This definition encompasses a range of situations including extensive and semi-extensive systems, and organic farming. In addition to arable and permanent crop systems, low-intensity livestock systems constitute a fundamental component of traditional land-use systems in Europe (Baldock *et al.*, 1994). Low-intensity or low-input livestock grazing farms contribute about one-third of all livestock grazing farms in the EU, cover roughly 15-25% of the European countryside (EEA, 2004), and constitute more than a 40% of the Utilized Agricultural Area (Andersen *et al.*, 2004).

In Europe, the origin of these systems is linked to the development of the traditional agricultural landscape after Mediaeval times (Plieninger *et al.*, 2006). After the mid-18th century, this landscape was substituted by industrial landscapes (Plieninger *et al.*, 2006), where intensive agriculture and high input systems became the norm. With industrialization and the Green Revolution, which prioritized agricultural production and food security for an increased human population, traditional farming was replaced by intensive agriculture in many areas. Only in marginal areas where intensification was difficult was traditional agriculture maintained, or agricultural activities abandoned.

In departing from traditional livestock systems, the general trend of intensification was a decoupling of animal and plant production systems and the intensification of both. The negative effects of intensified food production were the emergence of various acute environmental problems, such as P and N pollution (Berka *et al.*, 2001; Bleken *et al.*, 2005), and increased CO<sub>2</sub> and other greenhouse gas emissions, all resulting from the high dependency on non-renewable fertility and energy sources in intensive agricultural systems. On the other hand, intensification in animal farming led to forced animal feeding with cereals and other cellulose-poor foodstuff, making irrelevant the traditional concern for the grassland-herbivore interaction, and arguably creating problems for animal welfare. As a reaction, various social movements emerged whose common features were to stress the essential link between farming and nature, and to promote respect for natural equilibria (EC, 2001). By the late 1980s, the European Common Agricultural Policy (CAP) shifted from generating agricultural productivity gains, so as to make the European Community largely self-sufficient in its food supply, towards the promotion of quality products and the integration of environmental conservation into agriculture (EC, 2001).

### **Low-intensity livestock systems types in Europe**

Because of the wide variety of environmental and cultural conditions represented, low-intensity livestock systems in Europe are heterogeneous in structure, management and ownership, and include a wide range of habitats (Hopkins and Holz, 2006). In addition to sown and permanent grassland systems inside the farm, they include off-farm fodder systems such as extensive systems of communal pastures and traditional transhumance systems (Andersen *et al.*, 2004). Because of the intensification phenomenon, generally speaking, low-intensity livestock grazing farming is most dominant in unfertile regions in Europe, which helps explain why these systems are found primarily in upland and mountain areas, Mediterranean environments, wooded pastures, and more rarely in lowland areas. Structurally, these systems can be woodlands, and even forests or savanna-like ecosystems, but the most prominent low-intensity systems for livestock production in Europe are natural and semi-natural grasslands. Grasslands are ecosystems dominated by herbaceous species, mainly grasses, sedges and forbs. Among forbs, legumes play an important ecological role in terms of the nitrogen cycle (Jacot *et al.*, 2005), but can be negatively affected by grazing (Rochon *et al.*, 2004).

Plant traits vary according to the species selected by the environment, and there is an interaction between trait functional responses to grazing pressure and climate factors (de Bello *et al.*, 2005). Herb-dominated grasslands are usually more prominent in moist locations, while woody species increase in rangelands towards the dry Mediterranean regions (de Bello *et al.*, 2005). However, there are important exceptions to this generalization, such as ericoid shrub-dominated heathland in Atlantic climates, where the interaction between livestock grazing and other organisms is mediated by soil fertility, leading to complex vegetation dynamics between the heath and the grassland (Bokdam and Gleichman, 2000; Canals and Sebastià, 2002). In the Iberian Peninsula, an integrated agro-silvo-pastoral system was developed during the Middle Ages and expanded by the mid-20th century, termed *dehesas* in Spain, and also known as *montados* in Portugal (Díaz *et al.*, 1997). This particular rangeland system includes tree patches of differing density, and sometimes exhibits cultivated swards (Olea and San Miguel, 2006). Finally, in dryland areas of the Mediterranean basin, mixed-agro-pastoral systems with sheep rearing associated with cereal-fallow crops used to be very common, although this system is highly threatened (Correal *et al.*, 2006).

### **Direct benefits of low-intensity systems: livestock products**

Although in most areas dairy systems continue to intensify, meat production in sheep/beef tend to be low-intensity, reflecting poorer land quality or harsher climate. The survival of these latter systems often relies on niche market for organic or premium products, or if subsidies are provided to operate such a system. Many beef and sheep farmers are part-time farmers, often relying on agricultural subsidies not linked to output. However, in spite of the widespread perception that the main advantage of low-intensity livestock systems is related to conservation of the natural environment (Hopkins and Holz, 2006), these systems can be an important income source for farmers. Livestock product production can be maintained at good levels, sometimes after changes in management strategies (Porqueddu *et al.* 2005). Dairy cattle produced 6,000 kg milk animal<sup>-1</sup> year<sup>-1</sup> in nature reserves in the Netherlands, without any fertilizer application and with rotational grazing (Bakker and ter Heerdt, 2005). Milk from cows on fresh forage had much higher unsaturated:saturated fat proportion with more poly-unsaturated FA (beneficial for heart diseases) and more conjugated linoleic acid (possible anti-cancer effects) than milk from silage-fed cows, over a wide variety of regions, seasons, feeding systems and managements (Elgersma *et al.*, 2006). As a consequence, farmers from some dairy cooperatives in The Netherlands now receive a premium on top of their milk price (Elgersma *et al.*, 2006). Also, increased milk protein content was found in a Holstein dairy farm near Madrid after conversion from conventional to organic (Elvira *et al.*, 2007). The diet of ruminant animals can affect taste and the chemical composition of meat and dairy products with consequences for human health, and there is a niche for the marketing of food products by geographical origin, method of production, gastronomic value and nutritional and health properties (Hopkins and Holz, 2006). Another development opening the market for those products was the increasing public distrust of meat products originating in intensive farming systems, particularly after the detection of the Bovine Spongiform Encephalopathy syndrome (BSE). Products originating in low-intensity systems, including many farms in mountain and marginal areas, turned up to be safe and often organically-grown.

### **Direct non-commodity benefits of low-intensity systems: game hunting and other recreation activities**

In addition to livestock products, in the last decades a new important monetary benefit has emerged, linked to the aesthetic values and the wildlife richness of these systems, namely agro-tourism, outdoor activities and game-hunting. Agro-tourism and outdoor activities rely on the natural conservation values of low-intensity systems, and often provide the highest income in marginal agricultural areas (Hopkins and Holz, 2006), but this activity is not without risks, including increasing urbanization. Another important income source linked to the natural conservation values is game hunting. In the United Kingdom game hunting of birds is in some cases found to be extremely important to the rural economy (the Cobham report in 1992 suggested it to be worth around £700 million). In the Iberian Peninsula, in the last two decades many extensive livestock systems in unfertile, economically depressed Mediterranean areas have evolved into big (deer, wild boar, roe deer, fallow deer) and small game (rabbit, partridge, pigeon, turtledove) hunting farms (San Miguel, 2005). Particularly suitable regions can be found in the *dehesa* and *montado* systems, because these agro-silvo-pastoral ecosystems hold a rich plethora of flora and fauna, including game (Díaz *et al.*, 1997). Hunting activities now ensure more than 1.5 millions jobs and annually generate 1,800 million Euros in Spain (Federación Española de Caza, 2005), which indicates a high economic resurgence for otherwise depressed, low-productive areas. Nevertheless, the success and rapid growth of this extensive activity implies some serious threats for the preservation of the natural environment, with particular concern to the fragility of these ecosystems and their

high biodiversity (the majority are included in the Natura 2000 network). The danger therein lies not only in the actors' awareness or understanding of rapidly shifting land use preferences, but also the preparedness and experience of those responsible for mediating such changes. In this latter regard, threats mainly originate from: 1) the scarce existing tradition and knowledge of the handling of wild species in captivity, and 2) the general lack of management planning for such drastic shifts in land use. Game farms require: a) big spaces and vegetation mosaics (herbaceous and lignified) where animals may satisfy all their needs (shelter, food, fight, etc), b) very low stocking rates in order to avoid serious problems, such as confrontations and disease transmissions, and c) elevated quantities of digestible forage for every season, because wild animals are more selective than domestic stock and prefer browsing of shrubs than grazing of low-digestive grasslands, with its negative effects on less-tolerant lignified vegetation.

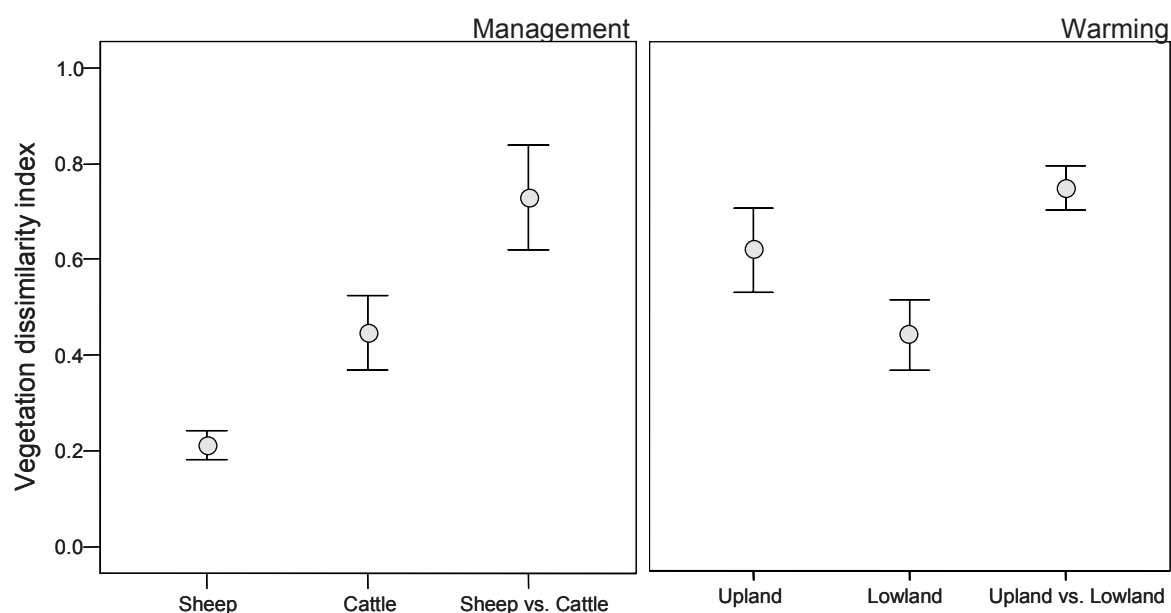


Figure 1. Changes in vegetation composition with changes in management (left) and climate (right). A vegetation similarity index was calculated, and the mean and 95% IC provided for plot comparison (within and between treatments). When the dissimilarity index is 0, there is the same vegetation in all plots; when it is 1, vegetation between plots is entirely different. Modified from Sebastià *et al.* (2008a) and Sebastià (2007).

### Indirect benefits of low-intensity systems: biodiversity conservation

One of the most highly valued and consistently cited benefits of low-intensity livestock systems is the maintenance of high biodiversity (Blanco *et al.*, 1998; Hopkins and Holz, 2007). More than 50% of Europe's most highly valued biotopes occur on low-intensity farmland (Bignal and MacCracken, 1996). Grassland biodiversity is both an externality of particular environments and farming systems, and also contributes to objectives of multi-functional land-use (Hopkins and Holz, 2006). Management can be used as a tool for multi-functional planning, including conservation and agricultural use (Austrheim and Eriksson, 2001), but a better link between ecological theory, applied research and practical applications is necessary. Interestingly, both abandonment and intensification have been cited as factors that threaten biodiversity in grassland (Hopkins and Holz, 2006). Abandonment alters plant diversity, and vegetation structure and composition (Milchunas and Lauenroth, 1993; de Bello



*et al.*, 2007). On the other hand, one effect of intensification is the lack of botanical and structural complexity in grasslands (McCracken and Tallowin, 2004). This negatively affects diversity and complexity across trophic webs, including bird (Wolff *et al.*, 2001) and arthropod (Di Giulio *et al.*, 2001; Kruess and Tscharntke, 2002) diversity, therefore impacting on ecosystem functions such as pollination, herbivory, predation and parasitism. However, plant species richness has been found to increase with grazing intensity (Cingolani *et al.*, 2005; de Bello *et al.*, 2006), suggesting a positive effect of disturbance on biodiversity in regularly disturbed systems such as grassland, likely until the pressure reaches a threshold at which system simplification occurs (Peter *et al.*, 2006). In addition, part of the diversity is due to the presence of common plant species, raising questions about biodiversity quantity versus quality (Canals and Sebastià, 2000; de Bello *et al.* 2006).

Although there has been much research on the effects on vegetation of grazing intensity (Altesor *et al.*, 2006; de Bello *et al.*, 2006) and abandonment (Biondini *et al.*, 1998; Frank, 2005), less is known about the sensitivity of grazed ecosystems to other grazing variables (Rook *et al.*, 2004, Bakker *et al.*, 2006). In a recent study, Sebastià *et al.* (2008a) found strong vegetation responses to changes in livestock type in paired plots across fences separating cattle-grazed and sheep-grazed mountain grasslands in the Pyrenees. Cattle-grazing favoured plant composition heterogeneity, while sheep-grazing homogenized the vegetation (Figure 1). Sheep showed a selective effect, but some of the selected species had special conservation value, including some endemics in the Pyrenees (Sebastià *et al.*, 2008a). On the other hand, increasing livestock-type diversity by mixing small (sheep) and big (cattle) livestock grazers could have additional benefits, and a positive effect of mixed grazing has been found on insectivorous passerine nesting (Evans *et al.*, 2006) and on soil carbon (Sebastià *et al.*, 2006).

### **Indirect benefits of low-intensity systems: ecosystem services**

Current ecological theory predicts that, with biodiversity loss, ecosystem functioning is threatened, as well as the goods and services that ecosystems provide (Chapin *et al.*, 1997, Hooper *et al.*, 2005), including those linked to soil and water quality, and climate change mitigation. Changes in environmental drivers can filter species (Sebastià *et al.*, 2008b) and guilds (Sebastià, 2007, Fig. 2), in turn affecting functional diversity (de Bello *et al.*, 2006) and the species' trait pool, including biomass allocation patterns (Sebastià, 2007). Tilman *et al.* (2006) found increases in the capacity for carbon sequestration with increased species richness in experimental systems, linked to increased belowground biomass allocation.

Land use changes and management regulate both biodiversity and ecosystem services provisioning. In Mediterranean ecosystems, livestock grazing management modifies vegetation structure in woodlands and can be used as a tool to control wild fire (Étienne *et al.*, 1995). Changes in plant community composition and diversity in response to changes in management and the environment have been reported to affect grassland functions such as herbage production (Kirwan *et al.*, 2007) and forage quality (Peter *et al.*, 2006; Sebastià, 2007). Conversion of cropland to grassland has been proposed as a measure to enhance soil C storage through increased production and SOM addition (Conant *et al.*, 2003), and reduce GHG emissions (Desjardins *et al.*, 2005). Importantly, the effect of management on soil carbon storage can be modified by other environmental factors, such as climate (Conant and Paustian, 2002) and topography (Garcia-Pausas *et al.*, 2007, Figure 3). Although soil organic carbon has been found to increase with grazing intensity (Conant *et al.*, 2001), this effect decreased with increased slope in Pyrenean grasslands, and the effect varied with climate (Figure 3). In addition, it is not clear what type of management will optimize the sequestration-emission balance, as there are many factors involved which have for the most part been studied separately. For example, N fertilization and the addition of N-fixing legumes have been listed as measures to enhance grassland productivity and therefore increase C sequestration

(Soussana and Lüscher, 2006; Hutchinson *et al.*, 2007), while recent studies (i.e., Garcia-Pausas *et al.*, unpublished) also find that N addition may enhance mineralization and, therefore, CO<sub>2</sub> emissions.

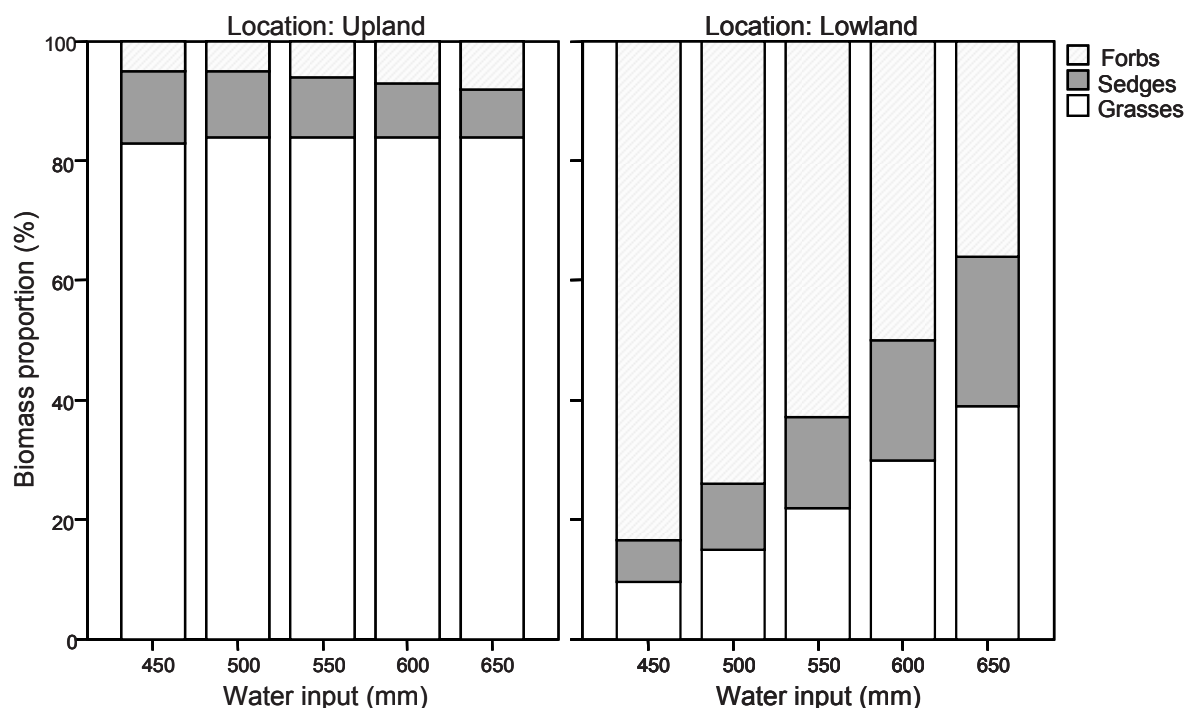


Figure 2. Strong shifts in plant guilds with water availability and warming in a transplanting experiment of grassland turves from subalpine locations (2000 m a.s.l.) to the lowland (700 m a.s.l.). Data from Sebastià (2007).

### Threats and challenges for management and conservation of low-intensity systems: land use and climate changes

Two important challenges threaten the conservation of low-intensity livestock systems in Europe: land use change and climate change. It has been shown above how grazing management can have a significant impact on the structure and function of grasslands and other low-intensity livestock systems. In the areas that are currently utilized for low-intensity agriculture, the main threats are abandonment, irregular distribution of grazing, and the loss of expertise in livestock management (Sebastià *et al.*, 2008a). Disentangling the effects of such land-use changes from the effects of climate change has become a central issue for predicting possible changes in goods and services that grassland ecosystems may provide in the near future.

Realistically, the greatest challenge for the near future is climate change. Recent findings show that in the last decade, emissions have increased at a higher rate than predicted by the most pessimistic scenario (Raupach *et al.*, 2007). Vegetation replacement was found in transplanting experiments in Swiss grassland after 7 years of warming (Bruehlheide, 2003). Strong responses of vegetation and biogeochemical properties were found over a single growing season in transplanting experiments of grassland in the Pyrenees (Sebastià, 2007). The findings of enhanced productivity found there seem to be linked to increased organic matter mineralization with warming, and is expected to decrease over time (Sebastià, 2007). More difficult to assess is the long-term effect of the decrease in herbage quality with observed shifts from grass-dominated to forb-dominated communities found in the experiments (Figure 2; Sebastià, 2007).



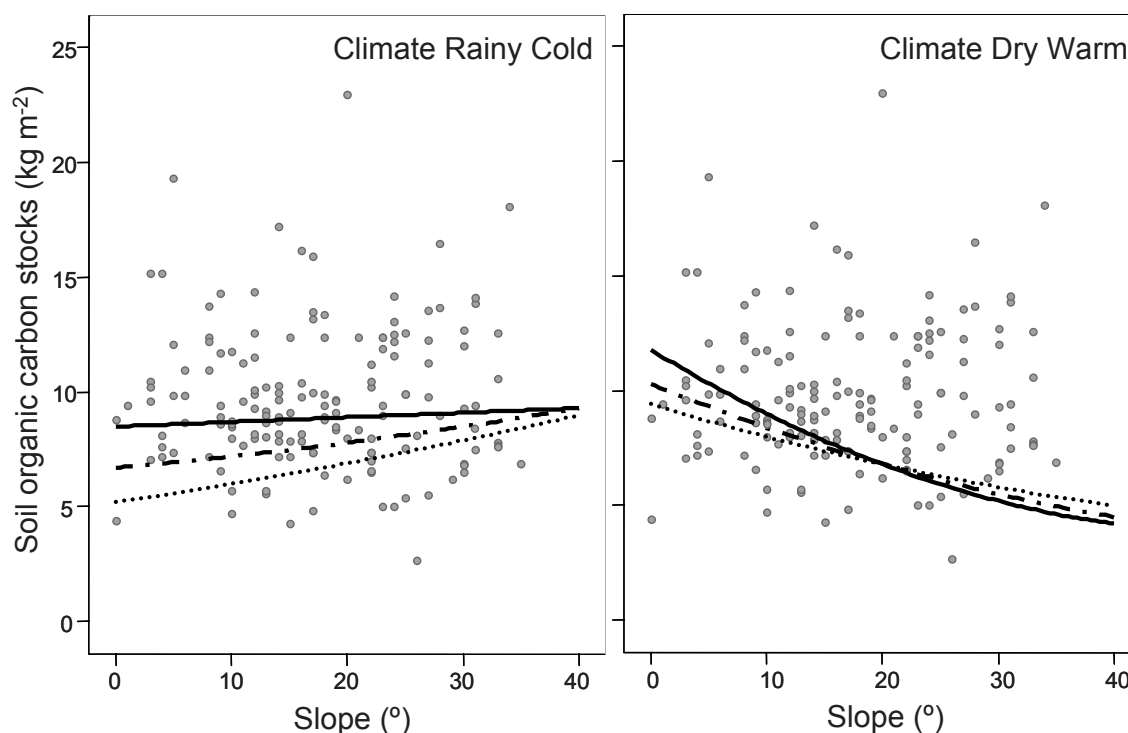


Figure 3. Changes in soil organic carbon stocks in the upper 20 cm soil layer (SOCS) with slope in two areas with different climate and under three different management regimes, where livestock are cattle at three grazing intensities: low grazing pressure ( $< 0.02 \text{ LU ha}^{-1}$ , dotted bar), moderate grazing pressure ( $0.2\text{-}0.4 \text{ LU ha}^{-1}$ , dashed bar), high grazing pressure ( $> 0.4 \text{ LU ha}^{-1}$ , solid bar). Climate rainy and cold: MAP 1300, MSP 450, MST-MST 5; Climate dry and warm: MAP 900, MSP 200, MST-MST 6. Where MAP is mean annual precipitation; MSP mean summer precipitation; and MST-MST mean annual temperature - mean summer temperature.

The foreseen changes are expected to lead to fundamental paradigmatic shifts in human societies. A substantial modification of attitudes and habits is necessary to face current environmental challenges. The response must be fast, including feeding habits, foodstuffs production and distribution, and consumer attitudes. The scenarios proposed by the Millenium Assessment (Alcamo *et al.*, 2005) are not enough to respond to the challenges posed, and more innovative measures are needed. Current scenarios must be solved, and new scenarios developed and implemented. These could come from deliberated and controlled choices and decisions (preferred), or will be imposed by reality from pressing external forces, coming too late and without planning. Within this new framework, low-intensity livestock systems are a highly viable option.

### Concluding remarks

Semi-natural grasslands and other rangelands are low-intensity systems used for livestock product production, usually extensively managed, grazed and/or cut, with no or low amounts of inorganic fertilizers, protection chemicals and fossil energy. Direct agro-environmental subsidies and decoupling from production subsidies within current European Common Agricultural Policy are favouring two livestock management systems: a) a very intensive one, linked to the market and disconnected from EU subsidies; and b) one that is more extensive and environmentally friendly, linked to quality product production, but dependent on EU subsidies (Rochon *et al.*, 2004; Cropper and Del Pozo-Ramos, 2006). Under current global

change conditions, there is pressing need for low-input, sustainable and environmentally-friendly systems. Low-input systems can offer health and ecosystem service benefits above and beyond their intensive counterparts; given the high performance in these areas, it is our view that low-input systems should not be relegated only to European marginal areas. Yet, key challenges remain, of which the most prominent may be rapid land use change in these systems, climate change, and long term economic viability and ecological sustainability. For both intensive and extensive systems, it is urgent and necessary to design innovative measures to face the new situation, within which low-intensity livestock systems are expected to have an increasingly important role.

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## **Session 5A**

### **The grassland landscape in Europe**



# Historical grazing pressure in south-central Sweden, 1620-1850

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## Abstract

Since biodiversity in semi-natural grasslands is largely a result of historical agricultural practices, knowledge about ecological aspects of former grassland management is essential in conservation biology. This study addresses the level of grazing pressure in four parishes of south-central Sweden. The pasture size (from cadastral maps, 1635-1850) was combined with livestock numbers (from livestock tax registers, 1620-1641, and probate inventories, 1750-1850) to reconstruct stocking density expressed as Grazing equivalents (Ge) ha<sup>-1</sup> pasture (1 Ge = maintenance demand of energy for 1 cattle, 0.7 horses, 4.8 sheep or 4.8 goats; all adult animals). During 1620-1641 the average stocking density in the four parishes was between 0.3 and 1.0 Ge ha<sup>-1</sup> pasture, but there were large fluctuations between years, and fluctuation could be used to calculate grazing pressure (proportion of available vegetation consumed). In average years 72-75% of the grazing resources were consumed. After the 17th century stocking density increased slightly in three parishes, implying either an increasing grazing pressure or an opening up of the tree-layer (enabling an increased fodder production per hectare) resulting in generally more intensive pasture use in the mid-19th century than in the 17th century.

Keywords: stocking density, grazing pressure, management history

## Introduction

In the pre-industrial agricultural Swedish landscape (*ca.* pre-1850) semi-natural grassland (unfertilized pastures and hay meadows) covered a large proportion of the countryside (Dahlström, 2006). Today only small fragments of grassland remain and considerable resources are used to preserve biodiversity in semi-natural grassland. This is to a large extent a legacy from the past (Gustavsson *et al.*, 2007), and management aiming to preserve grassland biodiversity should therefore benefit from knowledge about former management regimes. There is lack of knowledge about several aspects of historical management that may be of ecological significance. This paper focuses on the level of grazing pressure, changes in grazing pressure with time, and regional differences. Grazing pressure is defined as the fodder production/consumption-ratio within a given area (Hodgson, 1979).

## Materials and methods

Four parishes in south-central Sweden were chosen to represent both upland areas and lowlands. In the upland parishes (Alseda, the county of Jönköping and Kristberg, the county of Östergötland) agricultural production was historically divided between livestock production and grain production. In the lowland parishes (Fornåsa, the county of Östergötland and Selaön, the county of Södermanland) agriculture was dominated by grain production. The starting point of the studied period (1620-1850) was defined by the first usable historical sources, while the terminal point was set by the change in agricultural system. All calculations were made for each of the 61 individual hamlets. A complete description of the source material and methods is published in Dahlström (2006). A short summary is included here. First, the pasture area was extracted from cadastral maps from the 17th, 18th and 19th centuries. Secondly, livestock numbers were calculated from livestock tax registers (1620-1641)

and probate inventories (1775-1850). All grazing livestock were added into Grazing equivalents. (1 Grazing equivalent = 1 cattle unit = 0.7 horse units = 4.8 sheep units = 4.8 goat units. Each animal unit corresponds to the maintenance energy demand of adult animals). Livestock tax registers (1620-1641) allow a very exact and reliable calculation of livestock number for each farmer and for a succession of years. However, livestock under one year of age had to be added through other sources. From the variation in livestock numbers, the level of grazing pressure could be determined.

Probate inventories are lists of possessions of deceased persons. Since deaths of active farmers only occurred occasionally within each hamlet, inventories were assembled in 25-year periods in order to represent all farming units in the hamlets. This gives an approximate estimation of the livestock number, accurate enough to reveal differences in stocking densities between parishes and changes with time. However, since data were not always contemporary, livestock numbers were combined both with the closest preceding and the closest following data of pasture size, resulting in intervals of stocking densities. In cases of too few probate inventories, hamlets were excluded from a 25-year period.

## **Results and discussion**

### *Grazing pressure in the 17th century*

Within the period 1620 to 1641, livestock number varied with different amplitudes, the ratio: maximum / median livestock number being between 0.07 and 1.26 in the different hamlets. Mean amplitude in the four parishes was between 0.33 and 0.40. Irregularities in births and deaths in combination with a low number of animals caused most of the variation. To a smaller extent the variation could be explained by critical aspects of the sources. Using the assumption that maximum number of livestock entails a one-hundred percent consumption of available fodder, the deviation from the average number shows that the amount of unconsumed fodder in average years was 33-40 percent of the total. This gives an average grazing pressure of 72 to 75 percent. For single years the situation may differ considerably from the average grazing pressure. In years with fewer animals, the grazing pressure must have been lower, while years with many animals had a higher grazing pressure. However, extreme years may have been compensated with additional fodder, such as leaves. The variation may therefore be overestimated, and the average grazing pressure could have been even higher. On the other hand, the maximum grazing pressure may well have been lower than assumed here, leading to an overestimation of the average grazing pressure. In addition, different weather conditions (heat and precipitation) could have caused even larger fluctuations in grazing pressure, since weather conditions of the present year did not affect livestock number.

### *Regional differences*

There were no significant differences in grazing pressure between parishes (t-test), but the stocking densities were higher in the two lowland parishes (Fornåsa, Selaön) than in the upland parishes (Kristberg, Alseda (Table 1), with significant differences (t-test) between Fornåsa and both upland parishes and between Selaön and Alseda). This implies that there may have been differences in the quantity of available fodder. In Fornåsa, having the highest stocking density, the predominant bedrock types are sedimentary, and soil types are rich in lime (Fredén, 1994) giving a higher production capacity in general. The other lowland, Selaön, is dominated by acidic bedrock and pastures are generally situated on low productive till, the situation being similar to the two upland parishes. Soil properties can not fully explain the difference in stocking densities. Another factor influencing fodder production in grasslands is light influx, primarily determined by forest density. Higher stocking densities, in combination with equal grazing pressure, entails less densely forested pastures in lowlands

compared with the upland parishes. This conclusion is supported by qualitative descriptions of the forest resources in cadastral maps and other written documents, since descriptions of scarcity of wood was more common from the lowlands, especially from Selaön (Dahlström 2006).

Table 1. Average historical stocking densities in four Swedish parishes, calculated as Grazing equivalents  $\text{ha}^{-1}$  pasture. Min and max represents alternative calculations (see material and methods) of total livestock number (1620-1641) and pasture area (1775-1854). Standard error within brackets. N = number of studied hamlets.

Time period	Fornåsa, the county of Östergötland. N = 9		Selaön, the county of Södermanland. N = 27		Kristberg, the county of Östergötland. N = 10		Alseda, the county of Jönköping. N = 15	
	Min	Max	Min	Max	Min	Max	Min	Max
1620-1641	0.83 (0.12)	0.97 (0.15)	0.55 (0.04)	0.65 (0.05)	0.39 (0.11)	0.45 (0.13)	0.32 (0.05)	0.38 (0.05)
1775-1799	<i>0.87 (0.22)</i>	<i>0.94 (0.21)</i>	<i>0.60 (0.06)</i>	<i>0.61 (0.07)</i>	<i>0.44 (0.13)</i>	<i>0.45 (0.13)</i>	0.45 (0.09)	0.45 (0.09)
1800-1824	1.10 (0.19)	1.16 (0.16)	0.52 (0.05)	0.52 (0.06)	<i>0.37 (0.06)</i>	<i>0.40 (0.07)</i>	0.33 (0.03)	0.34 (0.04)
1825-1850	1.10 (0.20)	1.11 (0.20)	0.60 (0.06)	0.62 (0.07)	<i>0.40 (0.10)</i>	<i>0.47 (0.10)</i>	0.43 (0.07)	0.44 (0.07)
1849/1854	1.17 (0.14)	1.17 (0.14)	0.69 (0.06)	0.69 (0.06)			0.44 (0.05)	0.44 (0.05)

Note: 1 Grazing equivalent = 1 cattle unit = 0.7 horse units = 4.8 sheep units = 4.8 goat units. Each animal unit equals the maintenance energy demand of adult animals.

The pasture area includes pastures (grazed for five months), hay meadows (two months) and arable fields (one month after harvest). The area of hay meadows and arable fields were weighed according to the length of the grazing period.

Figures in italics denote periods with a reduced number of hamlets.

Sources: Livestock tax registers (1620-1641), probate inventories (1750-1850), cadastral maps (1636-1854).

### Changes with time

In all parishes, except Kristberg, the stocking density increased slightly between 1620 and 1850 (the increase being between 16 and 55 percent depending on the different calculations; Table 1). An increasing stocking density could possibly be associated with simultaneously increased grazing pressure, given a constant production of fodder. Inversely, a constant grazing pressure would be possible if the increase in stocking density was accompanied by an increase in fodder supply, created through less wooded and thereby lighter pastures. Neither change in grazing pressure nor in forest cover could be verified through analysis of qualitative information in maps and other written sources (Dahlström, 2006). Irrespective of which, the increase in stocking density implies an intensification of outland use during the studied 230-year period.

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# **Eighteenth century land use – a path to future conservation of grassland plant diversity**

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## **Abstract**

Historical management of semi-natural grasslands consisted of a variety of methods and activities. The current management, which in Sweden is almost exclusively full-season grazing, is therefore likely to be insufficient to successfully preserve the entire range of grassland plant diversity. In this paper we quantify the various grassland management regimes existing in an 18th century agricultural landscape in south-west Sweden and compare with their current land use. The results show that spatial habitat loss amounted to 92% of the original grassland area. Qualitatively, 58% of the grasslands still managed had experienced moderate to high changes in management regime. Historically, in the study area, six different methods of managing semi-natural grasslands were identified. In addition to the most common pasture with full-season grazing and hay meadow mown every-year, there were, e.g., pastures grazed in a two-year rhythm and alternating hay meadow and pasture. Today only full-season grazing is practised, irrespective of historical land use. Hence, in addition extensive habitat destruction, grassland plants also face a decrease in overall habitat quality. We suggest that reintroducing or imitating the historical management regimes of grasslands would increase conservation success.

Keywords: land-use history, management regime, historical map, grassland plant diversity, continuity, habitat quality

## **Introduction**

The drastic decline in grassland plant diversity is usually attributed to habitat loss, mainly due to cultivation, fertilization and abandonment (Stanners and Bourdeau, 1995). However, changes in management methods in the remaining grasslands may also contribute to the decline. Prior to the agricultural modernization in the 19th century, grasslands dominated the agricultural landscapes in large parts of Europe, not least in Scandinavia, where animal husbandry was important. The grasslands were managed with a variety of methods, of which full-season grazing and mid-July mowing were most widely used, but depending on the local agricultural system other regimes were also common (Gustavsson, 2007; Poschlod and Wallis De Vries, 2002). For example, low-productive grasslands enclosed with hay meadows and arable fields were not grazed until after harvest. The current grassland management, which in Sweden is normally grazing in May-October, is therefore most likely not a suitable management regime for all grassland plant species (Dahlström *et al.*, in press).

Historical grassland management regimes have been found to influence current grassland plant species richness (Bruun *et al.*, 2001). Gustavsson *et al.* (2007) even found the influence of land-use history to be more important than current land use, in particular 18th century management and the specific sequence by which one management regime changes or remains constant from the 18th to mid-20th century. One consequence of the importance of management regime is that broken regime-continuity reduces species richness. For example,

Gustavsson *et al.* (2007) showed that grazed grasslands with a long history of grazing had a higher mean number of grassland plant species than grazed grasslands with a meadow history. Moreover, changed management a century ago seemed to have reduced species number as much as did cessation of management some decades ago.

This study addresses the following questions: 1) what different 18th century grassland management regimes existed in the study area? 2) how are the grasslands managed today? 3) to what extent has the grassland management remained constant in the sites still managed?

## Materials and methods

The study was performed in 2005 in an agricultural landscape in Lidköping municipality, south-west Sweden (58°50'N, 12°91'E), covering approximately 1100 ha. Areas of 18th century semi-natural grassland and their management regimes were analysed from historical large-scale maps (Tollin, 1991). Current land use of the 18th century grasslands were analysed through field studies and the national database on semi-natural grasslands (Swedish Board of Agriculture, 2007), thereby quantifying habitat loss and changes in management regimes in grasslands still in use.

## Results

In the 18th century study area, semi-natural grassland covered *ca.* 652 hectares, i.e. about 60% of the land. Six different management regimes were found. Their proportional distribution is shown in Figure 1. Today, only 50 hectares are managed grassland – all as full-season pasture. The other 602 hectares have become arable fields (237 ha) or forest (365 ha) (Figure 1).

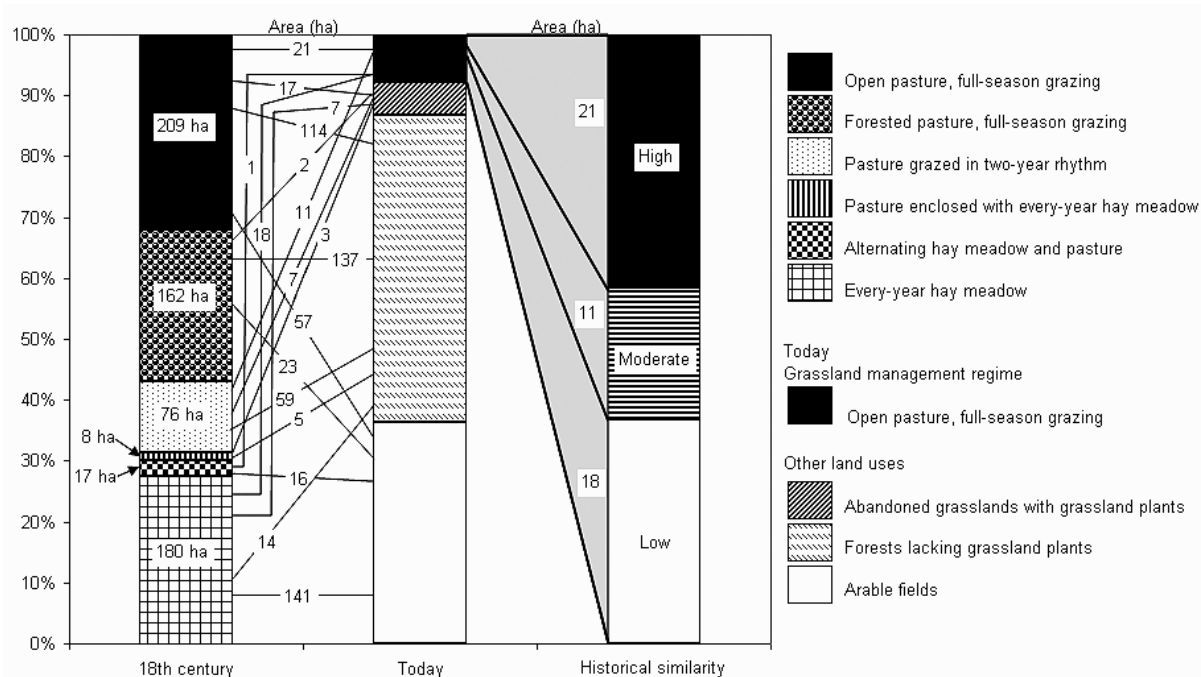


Figure 1. Area of 18th century grassland management regimes and their current use, both proportional distribution and actual area (652 ha total). Figures and lines between the columns show area subject to transition from one management regime to another. The right column shows the similarity in management regime in the currently grazed grasslands (50 ha) compared to their 18th century management regimes.

Thirty-six hectares of the forest still contained substantial populations of grassland plants, due to patchy canopy cover. The remaining 329 hectares contained no, or very few, grassland



plants, but consisted of modern production forest or dense successional forest. Among the currently grazed grasslands, 42% had also been grazed in the 18th century (Figure 1, high historical similarity); 22% were grazed historically, but in a two-year rhythm (moderate historical similarity) and the remaining 36% were mown historically (low historical similarity).

## Discussion

This study shows first, a serious decline in grassland area; secondly, a decline in the diversity of management regimes, and thirdly that 58% of the remaining currently grazed grassland in the study area has experienced considerable changes in management regime. The loss of grassland habitat is for the most part virtually irreversible. Only 5% of the original grassland area consists of successional forest that contains enough grassland plant populations to be worth restoration efforts. Gustavsson *et al.* (2007) did, however, find that these abandoned grasslands contained populations of specialized grassland plants, almost as abundant as in the grazed grasslands. Seeing as these former grasslands have not been grazed for the last few decades, they may contain grassland plants sensitive to full-season grazing, i.e. other species than those in the currently grazed grasslands. However, over a longer time perspective, these populations constitute an extinction debt.

The six management regimes that existed historically in this study area provided a wide variation in management timing, intensity and dynamics that favoured a larger range of grassland plant species than the current full-season grazing (Dahlström *et al.*, in press; Gustavsson, 2007). At the scale of a single grassland, switching the management regime is detrimental to some grassland plants (Maurer *et al.*, 2006). In a landscape perspective, the decrease in the diversity of management regimes implies a decrease in overall grassland habitat quality, i.e. a trivialization of the habitat. Hence, the habitat loss may be much more severe than the loss of grassland area, because of a serious decrease in habitat quality.

The experience of this project (Gustavsson, 2007) is that a greater variety in grassland management needs to be reintroduced, but that this variation should be based on site-specific land-use history. Reintroducing necessary components of the historical grassland management (c.f., Gustavsson, 2007) will not only benefit grassland plants, but also other organisms, which have been largely ignored in management practice (Söderström *et al.*, 2001).

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# **Agricultural grasslands in Bavaria – interrelationship of diversity and management**

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## **Abstract**

Changes in management practices and an overall loss of grassland areas have caused considerable changes in grassland structure and biodiversity. In particular, species-rich grassland types are exposed to the risk of extinction by intensification on the one hand, and abandonment on the other. To evaluate the current state of agricultural grasslands in Bavaria with regard to plant species richness, species composition and diversity, the Bavarian State Research Centre for Agriculture started a grassland vegetation survey in 2002. The survey focuses on grassland habitats of different levels of intensity and reflects regional differences in environmental conditions and management practices. More than 4400 relevés on plots of 25 m<sup>2</sup> have been recorded so far. Management intensity and type affect species richness significantly. Pastures, especially Alpine pastures, are more species rich than meadows. Average species number per plot is negatively correlated with stocking rate, and productivity is strongly correlated with management intensity. The survey shows a trend towards a general loss and uniformity of grasslands, and can be used as a tool to evaluate agri-environment schemes.

Keywords: grassland, biodiversity, survey, Bavaria

## **Introduction**

Agricultural grasslands are not only an important source of animal feed but also play a role as habitats. In Germany about 400 plant species are specialized on grassland (Korneck and Sukopp, 1988). Whereas the less intensive grassland management of former centuries increased biodiversity and led to the development of distinct grassland plant communities, restructuring and intensification of the last few decades has caused the opposite effect. In Bavaria, permanent grassland dropped from 43% (in 1960) to 36% of the agricultural land (Bayerisches LfU, 2003). Less productive fields are abandoned, and others are converted to arable land. The management practices changed and intensity often increased. The aim of the Bavarian grassland survey is to investigate the current state of the agricultural grassland.

## **Materials and methods**

The study was conducted in the federal state of Bavaria in the South of Germany. Studied grassland types vary from semi-natural Alpine pastures to improved permanent or sown swards. Permanent plots were chosen corresponding to the proportion of grassland area of the management types meadow and pasture. Each plot had a size of 25 m<sup>2</sup> and was marked permanently by a magnet. A plant list, the percentage biomass per species, the estimated total yield and the GPS coordinates of the plot were recorded. In addition information on livestock units per hectare (LU ha<sup>-1</sup>), management type and participation in Agri-environment-schemes (AES) were obtained from the agricultural administration. The first investigation cycle runs from 2002 to 2008. The presented data cover about 4,400 vegetation relevés from the years 2002 to 2006. LU ha<sup>-1</sup> was used as a surrogate for management intensity. To analyse the

interrelation of species richness, species composition and management, the plots were grouped into natural landscape units (Meynen and Schmithüsen, 1953) and means were calculated. To compare the species compositions, natural landscape units were grouped into eight greater regions (Figure 1) and the Sorensen index ( $C_S$ ) was calculated as:

$$C_S = 2j / (a + b),$$

where 'j' is the number of species shared by both groups, 'a' is the number of species of group 'A', and 'b' the number of species in group 'B'. The percentage biomass of *Rumex obtusifolius*, *R. crispus*, *Taraxacum officinale*, *Poa trivialis* and *Elymus repens* was summed to create the group 'weeds'. *Lolium* spp., *Trifolium repens*, *Poa pratensis* and *Dactylis glomerata* were combined into the group 'sown'.

## Results

The total number of species found in all the grassland plots totalled 730. Individual plots contained between 5 and 58 species of higher plants, 20.2 species on average. The average species number per plot of the natural landscape units showed a strong correlation with the proportion of herbs and a negative correlation with total yield and LU ha<sup>-1</sup> (Table 1).

Table 1: Spearman Correlation Coefficients ( $P < 0.001$ ) of natural landscape units.

ns – not significant, R. obtu – *Rumex obtusifolius*.

	LU ha <sup>-1</sup>	yield	grasses	herbs	legumes	sown	weeds	R. obtu
species per plot	-0.74	-0.76	-0.55	0.76	ns	-0.66	-0.63	-0.45
LU ha <sup>-1</sup>		0.84	ns	-0.60	ns	0.55	0.62	0.45
Yield			0.44	-0.66	ns	0.58	0.61	0.34

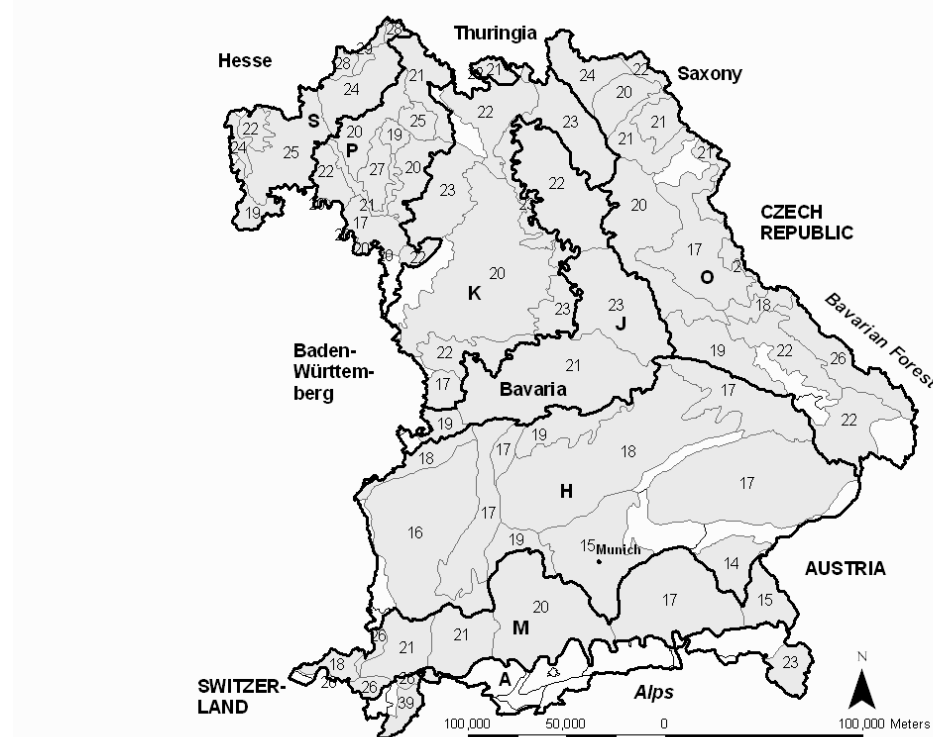


Figure 1: Map of Bavaria sub-divided into natural landscape units (grey, fine lines) and average number of species per 25m<sup>2</sup> plot. Natural landscape units not yet covered by the survey are displayed white. (dark, heavy lines: border of greater natural regions; capital letters: names of greater natural regions).

Average species number per plot decreased from 23.8 to 15.1 as stocking rates increased from  $< 0.5 \text{ LU ha}^{-1}$  to  $> 1.8 \text{ LU ha}^{-1}$ . There was relatively little variation in species richness and composition between plots (Figure 1). Even the most distant regions had, to a large extent, species in common (Table 2).

Table 2: Sorensen index comparing species composition of natural regions in Bavaria. (For geographical position of regions see Figure 1).

Region	A	M	H	O	J	K	P
M	0.65						
H	0.49	0.61					
O	0.58	0.64	0.73				
J	0.53	0.61	0.62	0.65			
K	0.50	0.61	0.67	0.68	0.73		
P	0.45	0.53	0.64	0.58	0.65	0.68	
S	0.55	0.64	0.67	0.72	0.64	0.71	0.65

## Discussion and conclusions

Even though a high total species number was found over all plots ( $\gamma$ -diversity), species richness ( $\alpha$ -diversity) was only moderate-to-low compared with mean species numbers in grasslands of other countries (Switzerland: 27-49 species per  $10 \text{ m}^2$  plot (Koordinationsstelle Biodiversitätsmonitoring Schweiz, 2006); GB: 22 species per  $200 \text{ m}^2$  (Carey *et al.*, 2002)). The distinct uniformity of species richness and composition in the different natural landscape units across Bavaria may be due to today's uniform management practices on intensive swards. In particular, continuous reseeding of highly productive grasses such as *Lolium perenne* may have furthered this conformity. An undesirable effect of increasing intensity is the increased biomass proportion of weeds as *Rumex obtusifolius*. Only natural landscape units in mountainous habitats, as in the Alps or the Bavarian Forest, where grassland is still used as extensive mountain pasture, had higher species numbers per plot (Figure 1). Concluding from our grassland survey, the agricultural grassland in Bavaria for the most part is species poor and shows a high conformity in species composition. As plant species diversity greatly depends on management intensity (Table 1), restrictions on stocking rates as supported by agri-environment schemes could enhance species richness and increase especially the proportion of herbs.

## Acknowledgements

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# Effects of summer drought on temperate grassland performance

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## Abstract

Climate models predict that summers in temperate regions will be drier in the future. The prognosis for Switzerland is for about 20% less summer precipitation in 2050. Since about a quarter of the Swiss area is covered by grasslands, we need detailed information about the consequences of such summer droughts to be able to develop adaptation strategies that ensure ecosystem services. Using transparent rain-out shelters, we simulated pronounced summer droughts in three Swiss grasslands at different altitudes and assessed the impacts on productivity, community structure and plant ecophysiology. We found that the reaction to drought was strongly dependent on grassland type. While reducing precipitation created water stress for the alpine grassland site, the effects were less clear in pre-alpine and lowland grasslands. At the pre-alpine site, a positive effect of drought on biomass productivity was observed. At the lowland site, grasses were more affected by drought than forbs, leading to a change in competitive interactions between plant functional types, favouring the weed species *Rumex obtusifolius*. Thus, our results indicate relevant consequences of climate change for grassland management, particularly in terms of productivity and weed pressure.

Keywords: climate change, drought, biomass productivity, weeds, temperate grassland

## Introduction

The extraordinarily dry summer in 2003 caused losses in grassland yield up to 65% in some regions of Switzerland (Keller and Fuhrer, 2004). Political actions had to be taken to react to a fodder shortcoming, e.g., cutting dates were shifted and the import taxes on hay were reduced (Bundesamt für Landwirtschaft BLW, 2003a, b). As Switzerland holds large water reserves in the Alps, drought has not been considered a problem for a long time. Additionally, due to the topographic diversity of the country, drought might be a regional rather than a national problem (Schorer, 2000). However, climate models predict that the frequency of extremely dry summers will increase in the future (Cubasch *et al.*, 1995; Schär *et al.*, 2004; Frei *et al.*, 2006). Simulations for northern Switzerland showed that precipitation between June and August will decrease to around 80% of today's values by 2050 (OcCC, 2007). The consequences such a decrease in precipitation has on temperate grassland ecosystems have not been studied very much and are not very well understood, yet. To be able to develop adaptation strategies and act in advance we need a better understanding of the reaction of grasslands to drought.

We therefore set up a drought experiment in three Swiss grasslands and investigated the effects of extreme summer droughts on the ecosystem. We hypothesised that drought would decrease above-ground biomass productivity of Swiss grasslands. However, different grasslands might be variously vulnerable to drier conditions.

## Materials and methods

The study was conducted in three grasslands at different altitudes across Switzerland: Alp Weissenstein (alpine, 1900 m a.s.l., Bergün GR), Frübüel (pre-alpine, 980 m a.s.l., Walchwil ZG) and Chamau (lowlands, 400 m a.s.l., Hünenberg ZG). These three grasslands are representative for Swiss agriculture at the respective altitude. In June 2005, we installed five trans-

parent rain-out shelters (3 x 3.5 m base area and 2.1 m high) at each of the three sites. In 2006, we added another shelter at Frübüel and two more at Chamau. During seven to eleven weeks in summer we excluded precipitation by covering the steel frames with transparent plastic foils, thereby producing a pronounced summer drought (Table 1). Control plots received the natural amount of rainfall.

Table 1. Duration of drought treatment and amount of precipitation excluded at the three grassland sites. Precipitation data (unpublished) provided by Rebecca Hiller<sup>a</sup> and Matthias J. Zeeman<sup>b</sup>.

	Alp Weissenstein	Frübüel	Chamau
Duration of drought (dates)	6.7.-25.8.2006	31.5.-17.8.2006	31.5.-17.8.2006
Precipitation excluded (mm)	234 <sup>a</sup>	371 <sup>b</sup>	254 <sup>b</sup>

Biomass was harvested at the respective cutting dates of the sites using fixed 20 x 50 cm collection frames. The cutting regime added up to one single cut at the alpine site, two cuts at the pre-alpine site, and six cuts at the lowland site. According to the common practice at the field sites, cutting height was approximately 7 cm. Biomass was separated into species, oven dried at 60 °C for one week and then weighed. Dry matter was used as an estimate of above-ground biomass productivity. Two biomass samples per plot were collected and pooled for analysis. Datasets were analysed using t-tests.

## Results and discussion

The effect of drought on biomass productivity depended on grassland type (Table 2). The only decrease in biomass productivity under drought was observed at the alpine site, as found in other studies (Tilman and El Haddi, 1992; Keller and Fuhrer, 2004; Ciais *et al.*, 2005). However, it is not unusual that rather wet grassland, typical for the Swiss pre-Alps, profits from drier conditions, since plant growth can be restricted by wet soils. Nevertheless, the regrowth of the vegetation was much slower under drought than under control conditions at the pre-alpine site. This resulted in no significant difference between biomass produced on drought plots compared to control plots at the second harvest after the treatment (data not shown), while we had observed a clear trend at the first harvest.

Table 2. Effect of drought on biomass yield (mean and standard error) at the three sites in 2006.

Biomass in g m <sup>-2</sup>	Alp Weissenstein (alpine)			Frübüel (pre-alpine)			Chamau (lowlands) <sup>a</sup>		
	Control	Drought	<i>P</i>	Control	Drought	<i>P</i>	Control	Drought	<i>P</i>
Total above-ground	284 (9)	184 (26)	<b>0.02</b>	330 (18)	406 (29)	0.06	271 (33)	353 (69)	ns
Total green	180 (11)	55 (7)	<b>&lt; 0.001</b>	297 (14)	349 (24)	0.10	264 (33)	331 (70)	ns
Grasses	129 (10)	36 (3)	<b>&lt; 0.001</b>	223 (18)	299 (34)	0.09	146 (34)	99 (18)	ns
Forbs <sup>b</sup>	47 (12)	19 (6)	0.08	66 (18)	46 (15)	ns	72 (29)	168 (77)	ns
Legumes	3 (0.5)	0.4 (0.2)	<b>0.002</b>	8 (4)	4 (2)	ns	45 (22)	64 (22)	ns
Total dead	104 (2)	128 (24)	ns	33 (7)	57 (10)	0.08	7 (1)	22 (7)	0.07

ns *P* > 0.1. <sup>a</sup>Sum of the two harvests that fully grew under drought conditions. <sup>b</sup>Including *Rumex obtusifolius*.

The trend towards more biomass produced under drought compared to control conditions at the lowland site was caused by the weed species *Rumex obtusifolius* L. increasing biomass production under drought conditions (Figure 1). In addition, photosynthesis of *Rumex* plants was much less reduced by drought than that of grasses (C. Signarbieux, pers. comm.). This resulted in a competitive advantage for *Rumex* over the grasses under drought, as reflected in



the grass biomass being slightly lower under drought. Although there was no significant reduction of total biomass due to drought, the amount and quality of the agricultural yield decreased due to the high contribution of *Rumex*.

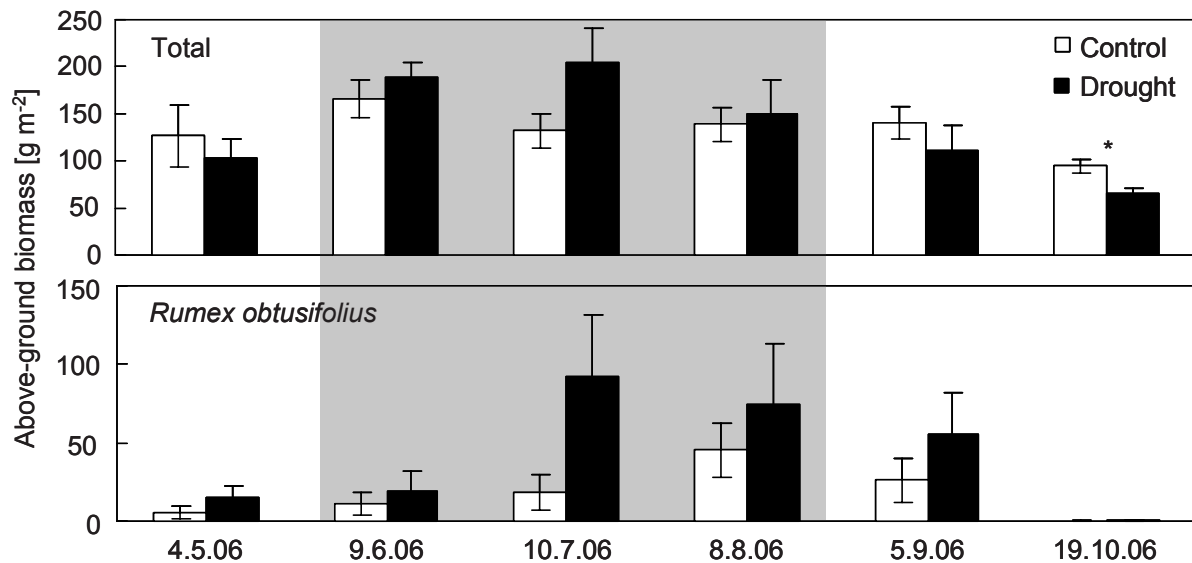


Figure 1. Mean dry matter yields (in g m<sup>-2</sup>) of total above-ground and *Rumex obtusifolius* biomass harvested at the lowland site Chamau in 2006. The shaded area represents the period of the drought treatment, bars indicate standard errors. \*  $P < 0.05$ ,  $n = 5-7$ .

## Conclusions

Our results show that climate change and especially changing summer precipitation will have differing effects on grasslands at different altitudes in Switzerland. It should thus be considered to develop specific adaptation strategies for different grassland types. Increasing weed pressure should be one main focus of future adaptation.

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# Botanical composition of mat-grass (*Nardus stricta*) grassland communities

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## Abstract

Matgrass (*Nardus stricta*) is a perennial invasive species of low forage quality. In Bosnia and Herzegovina mat-grass communities occupy up to 40% of hilly mountains grasslands and therefore are of significant economic interest. *Nardus stricta* is a particularly predominant species of the mat-grass associations, comprising over 70% of the sward. The rest of the botanical composition of mat-grass associations usually consists of species with low quality or even of no agricultural value. As the yield and forage quality depend on botanical composition of the sward, the aim of the research described in this paper was the identification and evaluation the botanical composition of this type of grassland. The study took place at the Rostovo mountain, 1,100 m a.s.l.. The botanical composition of the sward was recorded by the Braun-Blanquet method and species quality according to a method of Sostaric-Pisacic and Kovacevic. Results obtained showed that the *Nardus stricta* association is poor in botanical diversity as well as in forage quality. Only 17 species were found, and *Nardus stricta* comprised more than 80% of the total grass cover. Only two species were of very good forage quality (*Trisetum flavescens* and *Plantago lanceolata*) but their contribution to the sward was extremely low.

Keywords: mat-grass, mat-grass association, botanical composition

## Introduction

*Nardus stricta* is a plant of small size (typically 10 to 25 cm in height) with very narrow leaves. Its extremely dense tussock growth form suppresses growth of many other plants, and for this reason mat-grass associations are poor in terms of botanical composition. Although native to Eastern Europe it has been naturalized in many areas due to its very modest demand on soil and climatic conditions. Its appearance in grassland is usually a sign of acid soils poor in nutrients. It is called a 'cold xerophit' and can withstand shortage of water. However, it can also be found on wetlands of various types (Kissling *et al.*, 2005) where it increases its abundance and dominance, resulting in further negative effects on native species diversity.

Bosnia's hilly and mountain grasslands are usually rich in species diversity due to soil and climatic conditions, and also due to extensive methods of management. In addition, there are also large areas covered by mat-grass communities (up to 40%), in which *Nardus stricta* is more or less dominating. This grassland community is characterized by a reduced number of species, low forage yield per unit area, as well as poor quality of forage (Batinica, 1977). The aim of this study was to determine the botanical composition of typical mat-grass communities and to classify the species found in terms of their forage value.

## Materials and methods

Botanical composition of the mat-grass association was observed at the mountain Rostovo (1,100 m a.s.l.) in mid-Bosnia. Species abundance was recorded in ten plots (1m<sup>2</sup>) per ha (1 ha in total) using the Braun-Blanquet method. For classification of species quality, the methods of Sostaric-Pisacic and Kovacevic (1974) were used, which include nine quality classes: excellent (Exc), very good (Vg), good (G), moderate (M), poor (P), worthless (W), depressive (Dp), noxious (N) and very noxious (VN). This classification distinguishes species

quality as fresh fodder and hay, which can depend on species abundance as well as on the altitude at which the grassland is situated.

## Results and discussion

The studied grassland site with the mat-grass association was quite homogenous in botanical composition. Mat-grass was predominant in all plots (5.5, Table 1). The estimated proportion of mat-grass was 80 to 95% (in the sward grass cover). In total there were only 17 species (in some plots only 6 to 8). Due to the very dense sod, mat-grass eliminates or excludes many other species from the sward. This grassland community contained reduced botanical composition compared with others (Lakusic, 1987). Apart from mat-grass, *Agrostis capillaris*, *Festuca supina*, *Luzula campestris*, *Hypericum maculatum* and *Potentilla erecta* were also frequent in the grass cover. These species are characterized as species showing sociological affinity to *Nardus stricta* in grasslands and often appear in abundance within this grassland type.

Table 1. Abundance, forage quality as green matter and as hay classification and presence in observed plots (as %).

Species	Abundance	Classification in green matter	Classification in hay	Proportion (as %) found in studied plots
<i>Agrostis capillaris</i>	1.2	G	G	60
<i>Calmagrostis varia</i>	+0.1	W	W	30
<i>Festuca rubra</i>	+0.1	G	G	30
<i>Festuca supina</i>	1.2	P	W	70
<i>Nardus stricta</i>	5.5	Dp	Dp	100
<i>Trisetum flavescens</i>	+0.1	Vg	Vg	30
<i>Luzula campestris</i>	1.1	P	P	60
<i>Calluna vulgaris</i>	+0.1	W	W	50
<i>Galium cruciata</i>	+0.1	P	W	70
<i>Genista sagittalis</i>	+0.1	Dp	N	40
<i>Hypericum maculatum</i>	1.3	P	W	80
<i>Plantago lanceolata</i>	+0.1	Exc	Vg	40
<i>Potentilla erecta</i>	+0.3	P	W	90
<i>Polygala vulgaris</i>	+0.1	M	P	30
<i>Veronica officinalis</i>	+0.1	W	W	40
<i>Vaccinium myrtillus</i>	+0.1	W	W	50
<i>Viola canina</i>	+0.1	P	W	50

Most of species present were of poor agricultural quality (*Festuca supina*, *Luzula campestris*, *Galium cruciata*, *Hypericum maculatum*, *Potentilla erecta* and *Viola canina*) or even worthless (Table 1) (*Calmagrostis varia*, *Calluna vulgaris*, *Veronica officinalis* and *Vaccinium myrtillus*), according to Sostaric-Pisacic and Kovacevic (1974). Some of the above named species of poor quality (when grazed) are even worthless if they are used as dry feed or hay (*Festuca supina*, *Galium cruciata*, *Potentilla erecta*, *Viola canina*). Only four species (*Agrostis capillaris*, *Festuca rubra*, *Trisetum flavescens* and *Plantago lanceolata*) are of good to very good forage quality. However, their presence in the grassland community was extremely low (estimated as less than 10%).

The mat-grass is considered as depressive (having some negative effect in animal feeding) by Sostaric-Pisacic and Kovacevic (1974) and even as being noxious by Callihan and Miller (1999), because it is a coarse-textured grass that is not palatable to most livestock.

## Conclusions

Observations of botanical composition of *Nardus stricta* communities showed that the mat-grass association is poor in botanical diversity, with only 17 recorded species. In the mat-grass association, the presence of *Nardus stricta* was recorded in plots at up to 95% of the herbage in the sward. Classification of forage value of the species found in a *Nardus stricta* association indicates that forage from this type of grassland can be classified as worthless, depressive or even noxious.

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# **Environmental characteristics of abandoned grasslands, production orientation and socio-economic types of farms in the pre-alpine region of Slovenia**

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## **Abstract**

Land abandonment and forest expansion are distinctive processes in rural areas of the pre-alpine region in north-east Slovenia. This is widely seen as the result of a combination of socio-economic and environmental factors, and the relationships between these are very complex and vary at a local level. The aim of this investigation was to determine the characteristics of some environmental variables of farm parcels that are facing land abandonment and forest expansion. The sample comprised 71 farms with abandoned grassland and pastures. Environmental characteristics were evaluated on 211 parcels according to selected variables. In addition, the production orientation, the production structure and the socio-economic types of the farms were analysed. The results showed that the parcels were mainly situated on steep slopes, above the limit of mechanization, where manual work is required. Mixed agricultural production predominated, with low livestock stocking densities. Five socio-economic farm types were analysed, of which part-time farms predominated. The results are significant for broader research on the causes and patterns of land abandonment, and important for future measures of forest expansion in the region.

Keywords: environmental characteristics, production orientation, grassland, Slovenia

## **Introduction**

In Slovenia there is a relatively low proportion of cultivated land, with only 33% of the surface covered by agricultural land and almost 56% is covered by forest. Permanent grassland occupies 60% of the Utilized Agricultural Area. Especially in the Slovenian mountain areas, forest expansion in the last 47 years has increased by 16% and is still increasing (Krajnc, 2003). Statistical data show that abandonment of agricultural land has been spreading throughout Slovenia, but that it is most intensive and widespread in the Alpine and pre-Alpine regions. According to Perpar (2002) other important reasons for the abandonment of farm activities in the Slovenian mountains are: young farmers do not consider farming to have good prospects, agricultural incomes are low, farms are too small and agricultural land is too widely spread, there are poor natural conditions for agriculture production, a deficiency of farm successors, and there is a low reputation for agriculture production in general. However, agricultural production, and particularly livestock breeding, still have a crucial role for preserving the sustained presence of the population in order to maintain the cultural landscape and biodiversity (Borec *et al.*, 2004; 2005). This paper presents results of a study of some environmental and socio-economic characteristics of farms that are regarded as the main causes of forest expansion in the north-eastern part of the Slovene pre-Alpine region.

## **Material and methods**

The study area was situated in north-east Slovenia (Pohorje region) included a total of 1410 family farms. The study comprised 140 farms of which 71 farms were facing land

abandonment and forest expansion. On 71 farms a total of 211 parcels of land with forest expansion were studied in terms of five environmental variables (Table 1). The parcels were recorded with the help of a Geographic Information System (software ESRI ArcView) and according to digital cadastre plans. For the processing of environmental variables, IDRISI for Windows (version 2.007) and a Digital elevation model of Slovenia were used. For the statistical analysis of environmental characteristics, and land use variables, basic statistics were applied. Analysis of variance (ANOVA) was used for estimating the difference between different socio-economic farm types regarding the area of forest expansion.

## Results and discussion

The average agricultural land size of the farms is 9.9 ha, or 34.3 ha including forest. On agricultural land grassland use with pastures and orchards predominates. The average size of land that had become overgrown on farms is 2 ha. On the 211 parcels with forest expansion, pastures and extensive grassland predominate. Table 1 shows the main environmental characteristics of the 211 land parcels studied.

Table 1. General environmental characteristics of parcels facing forest expansion.

	Min	Max	Mean	Standard Deviation
Surface (ha)	0.01	11.79	1.30	2.01
Elevation (m)	540.0	1125.9	756.39	121.95
Orientation (degrees)	11.15	338.50	167.63	65.39
Inclination (degrees)	6.82	26.36	15.88	4.51
Distance (m)	0.35	992.07	276.23	271.2

The surface areas of the parcels facing forest expansion vary from 0.01 ha to 11.79 ha. Most are located above 700 m elevation, where abandonment of agricultural land occurs most frequently in Slovenia (rapid decline of grazing livestock in the last centuries). The mean orientation of the parcels is 167.63°, which corresponds to a southerly orientation. The parcels facing forest expansion are mainly oriented to the South, South West and South East (favourable orientation) which indicates that agricultural use predominated in the past. Most of the land parcels (90.5%) are located on land with slopes greater than 10° (mean 15.88°), where the use of agricultural mechanization is limited and there is a greater requirement for manual work. The maximum distance from farm to parcel where forest expansion occurs is less than 1 km. This is not surprising since isolated farms with the land tenure in the enclosure are typical settlements in the region (Gams, 1984). The agricultural land in general is therefore located relatively near to the farm buildings. However, 95.7% of the parcels border on wooded areas. The most common soil type on the parcels studied (86%) is Dystric Cambisols, which is also in general typical of the grazing and forest soils.

In terms of agricultural production, it was found that more than three quarters of the farms (78.6%) with forest expansion on agricultural land carry out diversified production (large number of various crops and livestock, combined production). Only 21.4% of them are oriented only to grazing livestock production, but on average they do not support more than 3 LSU ha<sup>-1</sup> (the average in Slovenia is 5.9 LSU ha<sup>-1</sup>). The analysis of socio-economic characteristics (after Kovačič, 1996) of farms with forest expansion showed that more than half (54.9%) are part-time farms and only 33.8% are full-time farms. Living farms represent 2.8%, aged farms 4.2% and supplementary farms 4.2% of the studied farms. ANOVA was used to explain the differences between socio-economic farm types regarding areas of forest expansion (Table 2).

Table 2. ANOVA for estimating the differences between socio-economic farm types regarding areas of forest expansion.

Socio-economic farm types	N	Mean area (ha) of forest expansion	sd.	F	P
<i>Full-time farm</i> : all family members in the active period of life work only on their farm or are supported.	24	1.739	1.497		
<i>Part-time farm</i> : at least one of the productive active members of the family nucleus (i.e. farm-owner and his/her wife/husband, his successor and his/her wife/husband) works exclusively on the farm; at the same time, at least one of these members is employed off-farm.	39	1.454	2.294		
<i>Living farm</i> : agriculturally non-active farm, where all family members live on the farm, but the agricultural land is leased.	2	1.750	0.353	4.508	0.003
<i>Aged farm</i> : all family members are older than 64 years and still (at least some) work actively in the farm.	3	6.900	5.369		
<i>Supplementary farm</i> : the productive active members of family are employed off-farm; they work in the farm exclusively during their free time.	3	2.601	0.537		
<i>Total</i>	71	1.837	2.374		

The results of ANOVA show that there is a significant difference ( $P = 0.003$ ) between different socio-economic farm types concerning forest expansion, which is not surprising. The high standard deviations indicate a high dispersion of results. *Post-hoc* analysis revealed significant differences between full time and aged farms and part time and aged farms. The mean values show that aged farms have the largest mean-size areas of expanded forest (6.9 ha) which could be explained by a lack of labour. The reasons for greater forest expansion on the supplementary farms are most likely connected with the predomination of non-agricultural activities on these farms.

## Conclusion

The study of environmental characteristics of land parcels shows that the natural conditions of the region are not favourable for agricultural use. Agricultural land abandonment and forest expansion occur mainly on parcels situated on steep slopes, above the limit of mechanization use, where manual work has an important role. The outcome of basic socio-economic characteristics and land-use analysis of farms also show inconvenient conditions for agricultural production. Mixed agricultural production with fewer livestock units on farms predominates. The farm characteristics show diversified agricultural production, with a small share of full-time farms and a low livestock pressure on pastures. All established farm and parcel characteristics indicate that agricultural land abandonment and forest expansion will continue in the near future unless appropriate development measures are taken.

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## Dry grasslands of Switzerland: an overview

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### Abstract

Dry grasslands are being surveyed in Switzerland for the Swiss Federal Office for the Environment. The aim is to preserve and promote the sites of national importance within the framework of a biotope inventory. During 1996-2007, 23,712 ha of dry grassland were evaluated as of national importance. Implementation of this inventory is based on a sustainable form of agriculture. Land management subsidies provide farmers with the motivation for appropriate land use. Further information: [www.umwelt-schweiz.ch/tww](http://www.umwelt-schweiz.ch/tww)

Keywords: dry grassland, biodiversity, conservation, Switzerland, semi-natural, subsidies

### Introduction

Switzerland is dominated by grasslands. The pronounced topography of an alpine country impedes crop-based agriculture, and approximately three-quarters of the agricultural area is used as meadows and pastures. Within the mosaic of land-use types and intensities, dry grasslands represent the most species-rich vegetation types. Since WWII there has been a dramatic decline in the spatial extent of dry grasslands, which has triggered the need for a large scale inventory to serve as baseline information for decision making and management. The political mandate associated with the inventory, which was completed in 2007, was to outline sites of national importance and to facilitate conservation of dry grasslands in all regions of Switzerland. Simultaneously, the political body is developing strategies and regulations, which will be of crucial importance for the control of future land use.

### Materials and methods

Habitats covered in the inventory were restricted to dry, oligotrophic or slightly fertilized, coline-to-subalpine grasslands. These belong to a range of vegetation units such as *Salvio-Arrhenaterion*, *Mesobromion*, *Nardion*, *Festucion variaae*, and *Caricion ferrugineae*. In a first step, aerial photos were used to delineate potential sites. In the field campaigns that followed, candidate sites were assessed and validated, among others, with GPS-referenced vegetation descriptions (*relevés*). From the beginning care was taken to establish and apply reproducible mapping standards as well as objective evaluation criteria which are explained in detail in Eggenberg *et al.* (2001). In order to define sites of national importance, a team of experts assigned numerical values and calculation modes to a set of indicators, i.e. vegetation type, vegetation diversity, floristic potential, structural elements, level of aggregation and connectivity (Figure 1) according to predefined criteria. The combination of target attainment values for each criterion and the differential weights then rendered a numerical value for each site between 0 and 1. Given the directive to conserve dry grassland in all regions and altitudes of Switzerland, final evaluation of the importance values was also influenced by regional frequencies and altitude. Based on the calculations, the political authorities decided on a threshold value to assign the final attribute of 'object of national importance'. Only these objects are part of the political inventory and are eligible for management subsidies from the state.

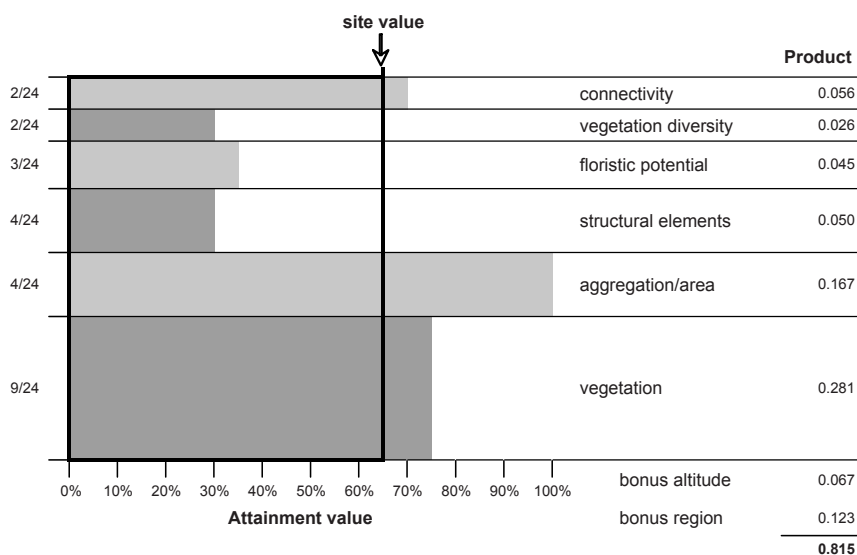


Figure 1. Calculation of the site value for a fictitious object (Eggenberg *et al.*, 2001, p. 224).

## Results and discussion

During 5,000 field days spread over 10 years, botanists visited 2,000 km<sup>2</sup> of grasslands and assessed over 13,000 distinct sites. In total, 23,700 ha of dry grasslands of national importance were identified (Figure 2) most of which were small in size (average size of 3.5 ha). Switzerland still possesses a diverse and functional network of dry grasslands in its mountainous regions (Jura, Pre-Alps and Alps). However, in the densely settled and intensively agriculturally used plains, only a few very isolated sites can be found (Figure 2).

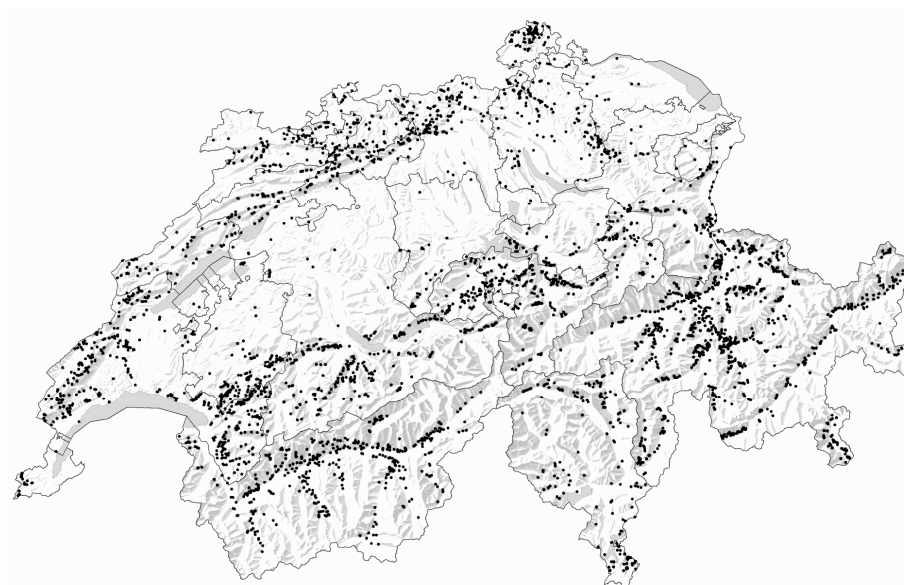


Figure 2. Distribution of dry grassland sites of national importance in Switzerland.

The total land-cover of dry grasslands comprises ~1.5% of the current Swiss agricultural area. One of the surprising results was the high number and area (~950 ha) of species rich 'natural' hay meadows (germ.: Wildheuplanken) in high mountain terrain. These largely hand-mown, labour-intensive meadows form part of the European alpine cultural heritage for which Switzerland has a special responsibility. Additional distribution data for all groups of organisms recorded in Swiss national databases were combined with the dry grassland inventory in order to identify hot spots of species diversity within the habitats being

considered (Figure 3). The primary data of the inventory as well as the different mapping products are prerequisites for adequate conservation planning. Additionally, the data set contains a lot of potential for continuous scientific research.

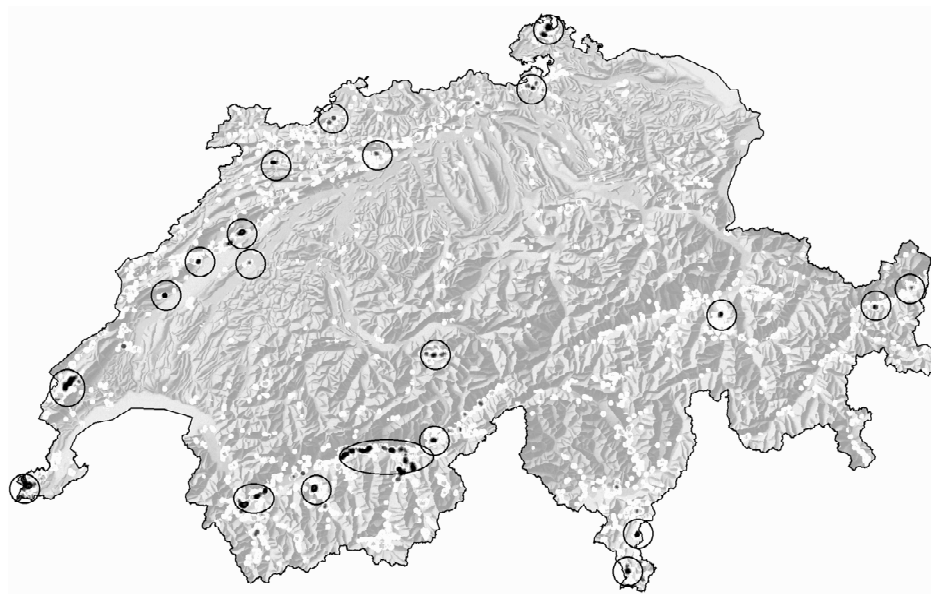


Figure 3. Species diversity hot spots for Swiss dry grasslands.

So far, the Swiss political bodies have relied on the inventory to implement a new ordinance that prohibits, amongst other things, fertilization and irrigation. As semi-natural grasslands, dry grasslands depend on a continuation of the traditional land-use practices. Therefore, contract-based governmental subsidies (20 M € per year) are given to farmers as an incentive for a sustainable land use. A newly developed 'priority region' category helps larger regions with the goal of protection of species that have higher demands on habitat area, e.g. butterflies and birds. To ensure conservation success in the future and to foster an understanding of the value of dry grasslands, government agencies put emphasis on information and training of active and practising farmers in the form of specific workshops, educational material, and close communication with the agricultural schools. In the light of increasing abandonment of dry grasslands, Swiss agricultural policies will have to adapt in order to conserve at least the most important sites. In this context, a number of innovative project have already been initiated (e.g. Landscape project Lake Biel: [www.vereinbielerseeschutz.ch/landschaftswerk](http://www.vereinbielerseeschutz.ch/landschaftswerk)).

## Conclusions

Effective conservation of dry grasslands requires a precise and standardized inventory. Based on 10 years of fieldwork, the 3,143 objects (23,700 ha) of national importance show that the mountainous regions of Switzerland support a relatively widespread network of species-rich dry grassland sites, whereas the plains are very poor. However, considerable financial incentives in form of governmental subsidies are needed to guarantee a continuous traditional management of dry grasslands.

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# Farmland bird indicators for evaluation of habitat quality in agricultural landscapes dominated by grassland areas

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## Abstract

Abundance is used for the purpose of characterization of habitat quality of landscapes for birds, and changes in bird abundances are considered to be sensitive indicators for habitat conditions. Abundance of selected bird species was recommended as indicators of biological diversity. Therefore, the aim is to develop a bird indicator based upon abundance in the agricultural landscape, with specific applicability for assessment of habitat quality in grassland dominated landscapes. The procedure was developed and tested in 2004-2006 in Germany, using 30 investigation areas which were randomly spread within grassland areas. For determining abundances the method of territory mapping was applied. Data were collected for 98 breeding bird species and 10 species were selected as indicators by statistical procedures. An abundance-based bird indicator was developed, and demonstrates the habitat conditions in landscapes, dominated by grassland areas from the perspective of species diversity. In this landscape type, habitat quality in time can only be moderately well classified. First analyses for evaluation of agri-environmental schemes by using indicator species resulted in slightly positive effects for territory density and species diversity. Improvement of measures and its long-term application are therefore recommended.

Keywords: farmland bird indicator, grassland landscape, breeding birds, abundance

## Introduction

The most important parameters used to characterize the suitability of habitats for birds are the occurrence and the abundance of breeding bird species, as a point of reference of the population density. For abundance, measurement unit in territories  $\times 10 \text{ ha}^{-1}$ , whose inquiry is carried out with the help of the territory mapping method, the population density of individual bird species or species associations in the different biotope structure complexes or landscapes can be determined. With knowledge of the spatial relationship between abundance and landscape, species populations can be calculated, while changes in the bird population are considered a sensitive indicator for the quality of the habitat conditions (Ten Brink, 2000). The dominating landscapes in central Europe are the very differently faceted agrarian landscapes, including grassland areas. Within the agrarian landscapes, the abundance of breeding bird species is closely related to the different types of agricultural land use on the cultivated areas and the parts and spatial distribution of natural, semi-natural and anthropogenic habitats. As a bird indicator to be developed for grassland-dominated agricultural landscapes, the abundance is used together with existing spatially and structurally differentiated features of agrarian landscapes as the initial parameter. For defined areas of landscapes, a methodical concept can be recommended for the representative field surveys of abundances, and be applied on larger areas. Based on the abundance measures and landscape data, for a selection of indicator bird species calculations are made of the local populations as well as extrapolations to estimate the meta-population of frequency and mean frequency breeding

birds. Ultimately, with the help of these results, a proposal can be made for a bird indicator for evaluations of the habitat qualities in agricultural landscapes dominated by grassland areas.

## Materials and methods

In Germany, the development of methods and their use is shown using as an example the federal state of Brandenburg (30,000 km<sup>2</sup>). The annual mean air temperature is about 8.5° C, and precipitation is about 600 mm. To census the abundance of single bird species, the territory mapping method was chosen. Data gathered on the territory are digitized in the context of biotope structure and area use and then entered in a GIS-supported database. The positioning of 30 investigation areas, each of 1 km<sup>2</sup>, was randomly distributed and stratified in the agrarian landscape, dominated by grassland areas. In order to ensure the above-indicated framework conditions for the positioning of the study areas, the systematization of the areas and their spatial borders was necessary. The total land area of Brandenburg was classified into spatially differentiated landscapes according to the appropriate biotopes (Hoffmann *et al.*, 2007), (Figure 1). The calculation and spatial marking off of the landscape types, as well as the agrarian landscape types, took place with the help of a GIS-supported calculation method using the Moving Windows Technology according to the dominance of existing biotopes. The field surveys took place in 2005 and 2006. On the basis of the field data, the abundance of the breeding bird species on the investigation areas and the mean abundance in the grassland areas on all sampled areas were calculated, using the size relation to the study areas and sizes of the complete agrarian landscape. As indicator species for the grassland areas, the selection of breeding bird species were defined, for which the habitats were exclusively or largely on open areas, and then indicator species were selected by the use of the Chi<sup>2</sup>-Test. Abundances and population sizes of these indicator species served as the basis for the bird indicators to be developed. For each indicator species, data on abundances were defined with the fitting population size, in which good, bad and intermediate habitat conditions of the agrarian landscape were found, and as a target value the abundance value for good habitat conditions was used. Attained target values serve as a measure for the classification of the abundance and population values of the indicator species found in the field surveys and for the calculation of the indicator.

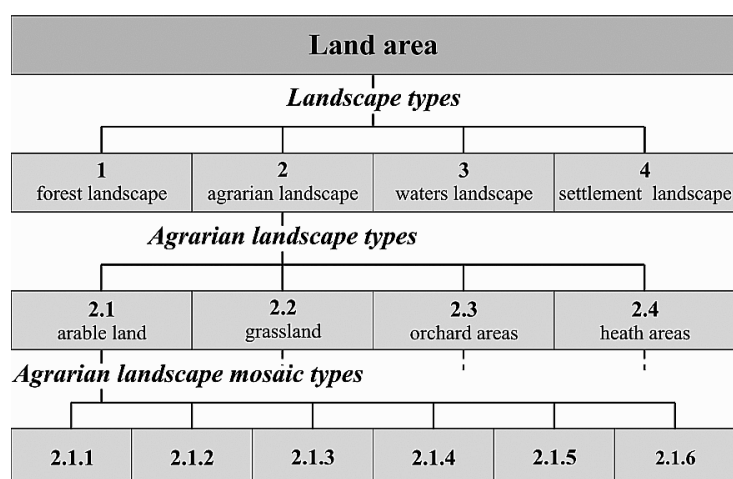


Figure 1. Model of landscape systematization.

## Results and discussion

With the help of the GIS-supported systematization and spatial marking off of the landscapes, the agrarian landscape was calculated for the positioning of the investigation areas.



The agrarian landscape covers 16,166 km<sup>2</sup> in total and within this the agrarian landscape type dominated by grassland areas covers 4,812 km<sup>2</sup>. In the two years studied, a total of 98 breeding bird species were identified. For each bird species, the size of the local population and the meta-population were calculated. In 2005/2006 the most frequent breeding bird species were *Alauda arvensis*, with 87,400/87,900 territories, and *Emberiza schoeniclus* with 33,500/30,300. From the total number of breeding birds, 10 species – *Alauda arvensis*, *Emberiza citrinella*, *Sylvia communis*, *Lanius collurio*, *Motacilla flava*, *Passer montanus*, *Anthus pratensis*, *Saxicola rubetra*, *Locustella naevia* and *Vanellus vanellus* – were identified as indicator species for the grassland areas. The abundance and population statistics for the indicator species, and also their defined target values, serve as the basis of information on bird indicators in grassland landscapes. The indicator is presented via the position of a pointer on a scaled circle. The placement of the pointer should indicate the situation of the habitat conditions for the indicator species in the landscape. Using these numerical values and additional colour scale from red to green with interim steps, habitat conditions from good (green) to bad (red) are defined. The number 100 is the target value for the indicator. Lower values amount to a successive deterioration of the habitat conditions. The calculated indicator (Figure 2) shows with 59.1% (2005) and 63.3% (2006) of the target value  $\geq 100\%$  habitat conditions for the grassland areas values that are neither good nor bad, but currently are relatively moderate. Analyses for evaluation of agri-environmental schemes by using the indicator species resulted in slightly positive effects for territory density and species diversity, but with different, partly contrary effects for the different indicator species.

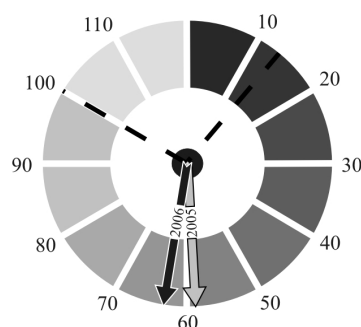


Figure 2. The farmland bird indicator 2005/2006 for the grassland landscape.

## Conclusions

Based on the habitat requirements of individual indicator species, recommendations for agricultural environment measures are possible as feedback to the practised methods of agricultural production and land use (Hoffmann and Greef, 2003). In addition to the actual indicator function as state indicator, it can be used as a measure for the improvement of existing agri-environment schemes from the nature conservation point of view. For this purpose, furthermore, information from individual investigation areas can be attained to show what presently serves as good habitats and land use forms for the bird species on agrarian landscapes through the favourable abundances shown with the indicator species.

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# Economic evaluation of the grasslands in two regional parks in Italy

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## Abstract

This paper describes a procedure for estimating the social benefits (option value and non-use values) to park visitors from a project for grassland conservation in two Regional Parks of the Bologna Apennines (Italy). A Contingent Valuation Method (CVM) survey by questionnaire was carried out in 2007. Respondents were asked their Willingness To Pay (WTP) per year for the conservation project. The main results show that the majority of respondents are in favour of the project implementation, and the mean WTP is about € 14 per year. Determinants of the WTP are income and being a member of associations for environmental goods conservation.

Keywords: grasslands, multi-functionality, option value, non-use value, contingent valuation method, regional parks

## Introduction

Over the last six decades, patterns of land cover have changed radically all over Europe, following two main tendencies: intensification and marginalization (Reger *et al.*, 2007). The hilly and mountain areas that have always been exploited with traditional non-intensive agro-pastoral systems have progressively been abandoned and naturally reforested. Conservation of mixed landscape dominated by grasslands appears more beneficial for biodiversity than other types of landscapes, such as homogeneous landscapes dominated by forests. Grasslands, moreover, are an interesting resource for recreation and tourism (Tempesta and Thiene, 2004) and, following the European Landscape Convention (2000), an important cultural and natural heritage. This paper aims to evaluate visitors' preference about a project for the conservation of grassland vegetation in the Italian parks of Monte Sole and of the Lakes of Suviana and Brasimone (Emilia-Romagna Region). The two parks are Sites of Community Importance and part of the Ecological Nature Network 2000. They are visited for their natural characteristics and for the cultural activities organized by the local communities. In these parks the conservation of grasslands is not profitable, and farmers are, in part, funded by the EU. The project considered here consists of the creation of a non-profit agency responsible for grassland conservation, and funded by private donations. Its cost is estimated to be about € 100,000 per year for the two parks.

## Materials and methods

### *The evaluation method*

We make reference to the concept of Total Economic Value (TEV) ascribed to grasslands, which is the sum of the use value, option value, and non-use values such as bequest value and existence value (Turner, 1999). We are interested in option value and non-use values. These are not established by the market and can be measured in monetary terms through the Contingent Valuation Method (CVM), which consists of a survey by questionnaire (Arrow *et al.*, 1993). A CVM survey was carried out in 2007 from May to August. Interviews were

made in the most visited areas of the two parks during the week-ends. The sample was chosen by interviewing 1 person in every 5 people met by interviewers in those areas. The evaluation questions are: i) 'Would you be willing to donate to a non-profit Agency something each year to cover the cost of the grassland conservation?' Photographs of two different landscape scenarios (grassland conservation and non-conservation) are shown and described to respondents in order to present the project. If respondents answer yes to question i), they are also asked: ii) 'What would be your maximum donation?'

## Results and discussion

The random sample consisted of 310 respondents, aged 18+. Incomplete observations (No. 7) and outliers (No.2) were removed. In these parks the main recreational activity is walking, and the second, cultural events. On a scale from 1 to 10, the mean score given to the parks as sites for recreational activities is 8.28. As regards environmental goods in general, visitors spend on average about € 32 per year, and about 30% of them are members of an association for environmental conservation. As regards other good causes, such as conservation of cultural goods and scientific research into cancer, the mean donation per year is about € 162.

Table 1. Determinants of the WTP (whole sample).

Models	Amemiya two-stage model		
Stage	Tobit model	i) Probit	ii) OLS
More Protection for Cultural Goods (MPCG)	17.956 (3.36) <sup>a</sup>	1.109 (4.15) <sup>a</sup>	6.910 (2.45) <sup>a</sup>
Member of Associations for the Conservation of Environmental Goods (MACEG)	9.021 (2.17)	0.944 (4.34)	
Yearly Expenditure for Environmental Goods (YEEG)	0.142 (3.61)		0.118 (5.38)
Yearly Donation for Other Good Causes (YDOGC)	0.033 (3.11)		0.020 (2.87)
Recreational Activity: Cultural Events (RA/CE)	11.253 (2.15)		8.265 (2.37)
High Rate to Park Recreational Activities (HRPRA)	10.007 (2.55)	0.602 (2.77)	5.131 (2.15)
Income	0.325 (2.86)		0.223 (3.04)
Missing Income		-0.459 (-2.21)	
Age (over 50)		-0.576 (-2.88)	
MR (Mill Ratio)			-8.733 (-3.16)
Constant	-36.762 (-5.89)	-1.039 (-3.50)	-2.248 (-0.62)

<sup>a</sup>t statistics for Tobit and OLS, and z statistics for Probit in brackets.

The great majority of respondents are generally in favour of more protection for environmental and cultural goods, and of grassland conservation in these parks; while 56% of respondents are willing to pay for their conservation. Zero values were not found to be outliers for regression diagnostics, and are not removed. Main non-donation motives are 'the State has to pay' (protest answer) and 'other projects are preferred'. For both parks together, the mean WTP is about € 14 (protest answers included), while for the Park of Monte Sole it is € 17, and for the Park of the Lakes of Suviana and Brasimone € 12. Main motives of donation are bequest value (57%), existence value (26%), and option value (15%). The number of park visitors is unknown, therefore we cannot estimate the aggregate WTP to be compared with the

total cost of the project per year. We can only say that, in order to cover this cost, at least 7,000 visitors should be willing to pay on average € 14 per year.

In order to theoretically validate the results of the CVM survey, since WTP values range from € 0 to € 100, two regression models were estimated: the Tobit model (Tobin, 1958); and the Amemiya (1978) two-stage model (first stage, Probit; and second stage, OLS). They are first estimated considering all the variables of interest, then stepwise estimations are made for  $\alpha = 0.05$  cut-off value (here presented). As regards income, the mean of each income category is considered; while the dummy 'missing income' represents those who do not state their income (45%). Table 1 shows that the Amemiya model (Probit, Pseudo  $R^2 = 0.2$ ; OLS, Adjusted  $R^2 = 0.3$ ) provides more information than the Tobit (Pseudo  $R^2 = 0.07$ ). The Mill Ratio coefficient is significant and shows that the decision to pay and that of how much to pay are dependent on and explained by different sets of variables. In both models, determinants of the WTP are MPCG and HRPRA. In particular, as regards the Tobit, giving a HRPRA increases the WTP; while as regards the Amemiya model, the Probit shows that it increases the probability to pay, and the OLS that it increases the WTP. Furthermore, as regards the Tobit and the Amemiya (second stage) models, if income increases, the WTP increases; in addition, spending for environmental goods (YEEG) and for other good causes (YDOGC), and attending cultural events (RA/CE) increases the WTP. The Tobit also shows that being MACEG increases the WTP; while the Amemiya (first stage) shows that being MACEG increases the probability to pay, and not stating income and being aged over 50 reduces it.

## Conclusions

The great majority of respondents are in favour of the project. The representative visitor is willing to pay about € 14 per year and rates the parks highly, is a member of an association for the conservation of environmental goods, believes in the need for more protection for cultural goods and attends cultural events in the parks. S/he donates for environmental goods in general, and for other good causes. When s/he states income, the higher the income, the higher the WTP; while when s/he does not state income, the probability to pay is lower.

## Acknowledgements

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# Variations in the phenology of semi-natural meadows in the western part of Switzerland

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## Abstract

The springtime phenology of more than 60 semi-natural meadows located between 400 m and 1200 m altitude was observed from 1995 to 2004. The goal of this study was (i) to validate a standardized method enabling the description of the developmental stage of meadows under various conditions and (ii) to provide farmers with references.

Each year, over a period of several weeks, the phenological stages of ten common species differing in their earliness (five grasses, one legume and four forbs) were monitored. Cocksfoot (*Dactylis glomerata*) was used as the reference species and the stages observed in other species were converted into 'equivalent cocksfoot stage' (ECS). The mean ECS, involving many species, allows the characterization of the stage of meadow development to be made on a standardized basis.

In the lowlands, the mean ECS 'full heading' was reached, on average, on 9 May and this stage was delayed by 4 days on average for each 100 m increase in altitude. Below 750 m, a difference of less than one stage between an early and a late year was observed for the same date. Above 1,000 m, this difference reached more than one and a half stages.

Keywords: semi-natural meadows, phenological development, altitude, temperature

## Introduction

Semi-natural meadows are generally composed of many species belonging to different functional groups. In addition to their proportion in the herbage, the stages of development of the herbaceous plants determine to a great extent the nutritive value of forage. Due to its heterogeneous botanical composition, it is sometimes difficult to determine the 'mean' stage of development of a meadow. Standardization of phenological data enables a precise comparison to be made of the developmental rhythm of different grasslands on a common basis.

The nutritive value of forage is usually estimated by means of practical tools that do not necessitate chemical analysis. The most commonly used tool takes into consideration the three main factors influencing forage quality: stage of development, type of botanical composition and conservation method (e.g. ADCF, 2006).

This paper describes the method used to characterize the phenological stage of meadows on a standardized basis. Some examples of practical applications are also given. For different thermal levels, the model gives, among other things, relationships between the phenological 'mean' stage and the Julian dates. This information is useful since few farmers in Switzerland are fully aware of a meadow's phenological stage at the time of the first harvest.

## Materials and methods

### *Calibration and development of the model*

Between 1990 and 1994, a model was calibrated describing the developmental rhythm of different species by comparing them to cocksfoot (= reference species). Ten species were chosen for the assessment of phenological development: five grasses, one legume and four

dicotyledonous plants (Table 1). These species are common, easy to recognize and present a broad variation of developmental rhythm (earliness). Two independent scales of notation were used: one for the grasses and another for the dicotyledonous plants (Table 2). Both of them are inspired by the work of Caputa and Sustar (1975) as well as Fleury (1983). The notations were made on semi-natural meadows located at ten different thermal levels, depending on altitude, vegetation period and mean temperature (Schreiber, 1977). None of the meadows presented characteristics of extreme conditions (e.g. slope or soil type). Every seven to ten days, at least ten shoots per species were observed from the stage 'flower buds' of the dandelion (*Taraxacum officinale*) up until the time of the first cut of the meadow. In the present model, relationships between the nine species and the cocksfoot were assumed to be linear (Table 1) and to remain unchanged in all places and every year.

Table 1. Species observed and equations used to calculate the equivalent cocksfoot stage (ECS).

Common name	Latin binomial	Family	Earliness	Approximate stage of the species when the full heading stage is reached by cocksfoot	Regression $x = \text{stage of the species, } y = \text{ECS}$
Cocksfoot	<i>Dactylis glomerata</i>	Poaceae	medium	4*	—
Meadow foxtail	<i>Alopecurus pratensis</i>	Poaceae	early	5.5	$y = 0.91 x - 0.9$
Smooth meadow grass	<i>Poa trivialis</i>	Poaceae	medium	4.5	$y = 0.88 x - 0.04$
Perennial ryegrass	<i>Lolium perenne</i>	Poaceae	medium	4.0	$y = x - 0.2$
Timothy	<i>Phleum pratense</i>	Poaceae	late	2.5	$y = 1.43 x + 0.3$
Dandelion	<i>Taraxacum officinale</i>	Asteraceae	early	7.0	$y = 0.67 x - 0.7$
Wild chervil	<i>Anthriscus sylvestris</i>	Apiaceae	medium	5.5	$y = x - 1.6$
Meadow buttercup	<i>Ranunculus acris</i>	Ranunculaceae	medium	5.0	$y = 0.95 x - 0.95$
Red clover	<i>Trifolium pratense</i>	Fabaceae	late	4.0	$y = 1.25 x - 1.1$
Oxeye daisy	<i>Leucanthemum vulgare</i>	Asteraceae	late	4.0	$y = 1.11 x - 0.3$

\*The cocksfoot is used as reference. Details of the phenological stages are given in Table 2.

Table 2. Stages of phenological development for the grasses and the dicotyledonous plants.

Grasses	Dicotyledonous plants
1. Tillering	1. Vegetative stage (most of the plants have developed three leaves)
2. Transition phase (elongation)	2. Flower buds (buds can be seen on more than 50% of plants)
3. Start of heading (10% of plants with ears)	3. Stem elongation (flower buds developed on more than 50% of the plants)
4. Full heading (50% of plants with ears)	4. Start of flowering (10% of plants with flowers)
5. End of heading (90% of plants with ears)	5. Full flowering (50% of plants with flowers)
6. Full flowering	6. End of flowering (90% of plants with flowers)
7. Fructification	7. Fructification
8. Dispersal	8. Dispersal

### Validation of the model

Between 1995 and 2004, over 60 semi-natural meadows located at different thermal levels were monitored by various people working in research and/or rural extension. The observation network included some surfaces that had already been used for calibration. Most of the 60 meadows were observed over periods covering several consecutive years. The notations were made in the same manner as for the calibration.

Taking the ECS of all species observed on a meadow at a given time as a starting point, the mean ECS was calculated. For practical reasons, the species data were not weighted by their herbage biomass. Finally, the ECS averages for the entire season were adjusted to Julian dates by means of a polynomial regression.

### Results and discussion

Equations for calculation of the ECS obtained from data collected between 1995 and 2004 were very similar to those from the calibration period. Variations in the realization date of the mean ECS = 4 between the thermal levels and the years are indicated in Figure 1. This stage



was delayed by 4 days on average for each altitude increase of 100 m. Variations between years were much greater in the mountains than in the lowlands. A difference of 8 days between an early and a late year was recorded in the lowlands, whereas up to 16 days' difference was measured in the highest areas. According to these observations, the stages of development change about every 11 days. In terms of nutritive value, a difference of one stage of development corresponds approximately to a decrease of about 3 points of digestibility (ADCF, 2006).

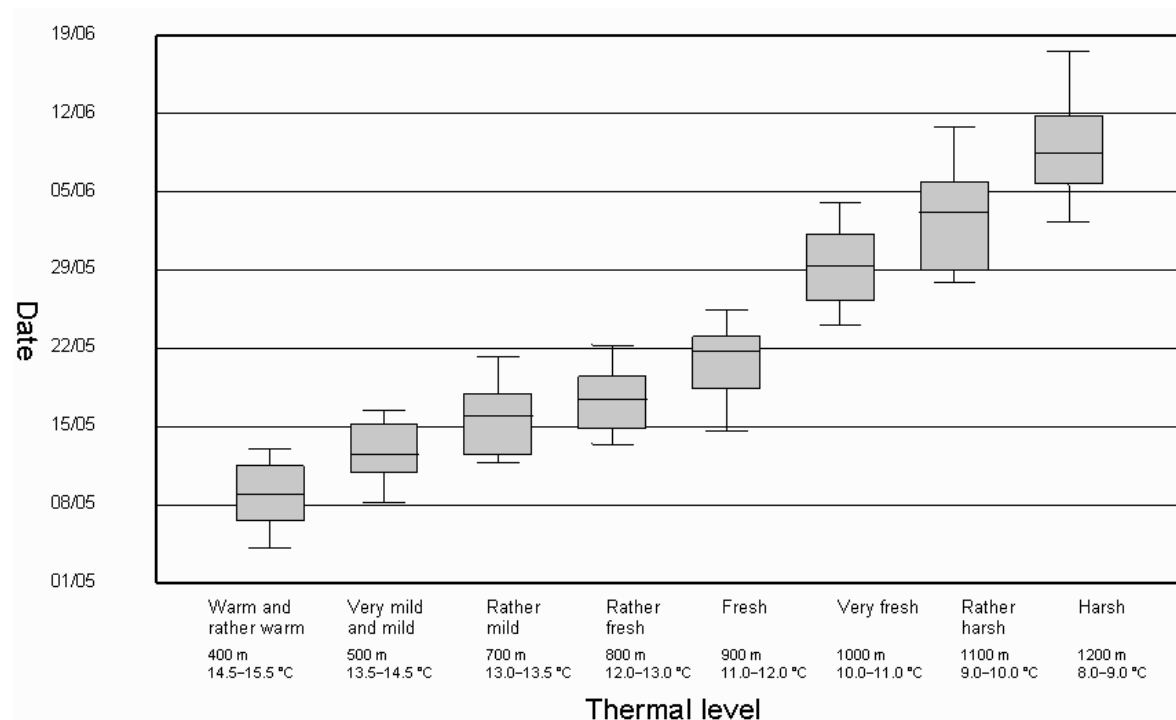


Figure 1. Variations in realization dates of equivalent cocksfoot stage 'full heading' (mean ECS = 4) for the different thermal levels, characterised by altitude and mean temperatures between April and October. Mean values for the period 1995 to 2004 (extreme values, first and third quartiles, median).

## Conclusions

Despite some limitations, the method presented in this study helps in describing the phenological development of semi-natural meadows and consequently leads to a more accurate assessment of forage quality. Due to its simplicity, this model has met with success and been widely used by rural extension since 1995 in the western part of Switzerland.

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## Landscape values of grasslands

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### Abstract

Growing multifunctional demands on grasslands have increased interest in landscape values of grasslands. Many scientific publications have highlighted the importance of grasslands in landscape architecture, but relatively less has been published about the landscape values of grasslands. We discuss, based on personal experiences (in research and education), local values (openness, canopy structure, heterogeneity, colours and shades, continuity, relation to other landscape elements) and landscape values of grassland. Among amenity functions, it is predicted that the landscape values of grasslands in developed countries will increase.

### Introduction

Grasslands and rangeland ecosystems constitute vast natural resources. After forests, they are the second most common terrestrial ecosystems, with a total area approximately twice that of croplands and half that of forests.

On a continental scale, grasslands are the most common land use systems, except in Europe, where both croplands and forests occupy a higher proportion of the land-use area than grasslands. A recent review of multifunctional demands on grasslands (Nagy, 2007) presents a versatile picture of grassland products (material goods/commodity outputs) and services (non-material goods/non-commodity outputs). Among these benefits, aesthetic or landscape values are especially important (Carlier *et al.*, 2005; Herzog *et al.*, 2005; Nagy, 2005; Vella *et al.*, 2005) in terms of stressing the importance of grasslands in landscape design and development. However, until now, little has been published on the details of the landscape values of grasslands.

A landscape is a visual and aesthetic phenomenon; a way in which visible elements of a given area (the objective 'picture') are reflected and perceived in the human mind (the subjective 'evaluation'). Objective elements include, e.g., spatial dimensions, forms, colours and materials. The subjective elements may refer to the individual appreciation of a set of landscape elements (e.g., monotonic, heterogeneous or harmonic landscapes) and/or the aesthetic qualification of given landscapes (e.g. nice, beautiful, attractive, boring, interesting).

Beyond the subjective nature of landscapes, there are methodological difficulties influencing landscape evaluation. One of these is the distance from which a landscape is experienced. From long distances, only large-scale patterns can be evaluated. From shorter distances, one can evaluate more detailed structures.

In this paper we discuss:

- methodological difficulties in defining landscape features,
- local features of grasslands (originating from grassland vegetation itself),
- landscape features of grasslands (relation to other landscape elements, e.g. natural or anthropogenic),
- importance of grasslands in landscapes appreciated by humans

### Local features of grasslands

A major landscape value associated with grasslands is openness. Open landscapes are associated with freedom, making it possible to see long distances, to identify remote objects and offer habitats for fauna, which require open landscapes (e.g. ground nesting and raptorial birds). At short distances, high vegetation (e.g. in savannas), may reduce the openness of the landscape.

Homogeneity or heterogeneity is a second feature of grasslands. From a distance, grasslands appear to be homogeneous, natural landscape elements. However, at short distances, homogeneity is replaced by versatile heterogeneity regarding sward surface, structure, colour or density.

The green colour of a grassland landscape reminds people of nature and living vegetation in general. Depending on local conditions, a wide range of greens, in a variety of shades and more or less homogenous in colour can be seen. From short distances, however, one can sometimes recognize many colours and shades on grassland originating from the different colours of grasses (e.g. flowering spikes, panicles, dried material) or flowering herbs in species-rich, permanent grasslands.

The structure of the grass canopy creates a positive image in our mind.

Another important feature of grasslands is the long continuity of these landscape elements. The huge worldwide grassland ecosystems (the Eurasian steppe, the savannas in Africa, the Pampas of South America, and the prairies of North America) form historical landscapes. In Europe, the grassland landscape may belong to the identity of a country (hill pastures in Switzerland) or of a region (*dehesa* in Spain and Portugal) and have constituted its image for outsiders for ages.

### Landscape features of grasslands

Grassland ecosystems include specific wildlife and/or grazing domestic animals. Visible animals, whether wild or domestic, make a grassland landscape more attractive. Not only these animals, but also historical anthropogenic landscape elements are important elements in grassland landscapes, e.g. sweep-pole wells on the Hortobágy in Hungary, the haystack poles in mountain meadows in Transylvania, in humid meadows in Poland, and parts of Scandinavia, slate paddock fences in the Scottish highlands or wooden windmills in Holland, are all integral landscape elements in these areas. In contrast, modern landscape elements (e.g. steel towers for wind power stations) are rejected in the traditional grassland landscapes. In Hungary, external landscape features of grasslands are vital on nature reservation areas, where preservation of historical landscapes is of key importance. Landscape objects made of natural materials (e.g. wood, clay-brick, and reed) which follow traditional local architecture styles are welcomed by nature preservation agencies.

Grass vegetation is also commonly used in man-made landscapes. On urban, suburban and rural road verges, motorway embankments, dam faces, railway embankments etc., artificial grasslands are most commonly used. In urban parks, open spaces and domestic lawns, man-made turf is able to harmonize with any artificial landscape object (e.g. concrete surfaces, buildings, ornamental plants), playing an outstanding role in social amenity.

In general, there is a worldwide interest in maintaining grasslands in historical landscapes. This interest is mostly connected with preserving ecosystems and habitats for wildlife. In modern societies, grasslands are increasingly used to meet amenity functions in landscapes. In several countries (e.g. Denmark – Gronbaek (2007); Hungary – Peregi and Füsti (2005), grass seed production and trade for amenity purposes has become greater than that for agriculture alone.

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# Influence of some climatic factors on invasive species coverage index in western Romania

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## Abstract

During the last two decades in Romania, and in other European countries, the number of the herbivores that are using grassland has decreased dramatically, leading to a poor management or abandonment of these surfaces and the spreading of invasive species. The purpose of this work is to analyse the influence of some climatic factors (air temperature, rainfall, air relative humidity and soil surface temperature) on the coverage index of invasive species in western Romania. For every model the correlation coefficient exceeds 0.8, showing an interrelation among the analysed variables, with a direct and moderate correlation. The species studied act as invasives, considered in terms of their spreading coefficient, except *Xanthium spinosum*.

Keywords: grassland, vegetation, climatic factors, invasive species

## Introduction

Previous research has demonstrated that climatic factors have a decisive role on the characteristics of vegetation cover. According to Maxwell *et al.* (2003), climate and the topography play important roles influencing plant species invasions. Knapp (2002) reported that in recent during the last decades the average temperature has increased and rainfall and its seasonal distribution are changing, and that climate models are predicting an increased frequency of climate extremes, such as large amounts of rainfall in a very short time followed by very severe droughts. Myers *et al.* (2004) suggested that it is possible to realize predictions concerning the invasion of species that are responding to temperature and rainfall changes along with habitat disturbance. Zedler (2004) proposed that invasive species have greater tolerance limits for the environmental factors, thus tolerating extreme conditions better than non-invasive species. The objective of this study is to reveal the influence of climatic factors on the specific coverage index of some grassland species from the point of invasiveness.

## Materials and methods

This study was performed during 2003-2005 on 26 permanent grasslands situated in places with different environmental conditions from Banat region of western Romania. Data were collected once a year. The target species studied are from different botanical families and include herbaceous species (*Juncus effusus*, *Carduus acanthoides*, *Carlina vulgaris*, *Carthamus lanatus*, *Cirsium undulatum*, *Xanthium spinosum*, *Eryngium campestre*, *Dipsacus fullonum*, *Euphorbia cyparissias*, *Calamagrostis epigeios* and *Pteridium aquilinum*) and shrubs and sub-shrubs (*Prunus spinosa*, *Crataegus monogyna*, *Rosa canina*, *Rubus caesius*, *Sarothamnus scoparius*, *Amorpha fruticosa* and *Ononis spinosa*). The grasslands studied have different coverage of invasive species, and different vegetation features (dominance, biodiversity, etc.). This study was made at a local scale, within areas of limited infestation, and with a focus on the extent of the small patches covered with potentially invasive species. The units of the temporal scale are years (2003-2005). Plots were situated at altitudes between 87 and 370 m, on soils with pH range 5.4-8.0. The mapping of the aerial projection of plants on 100 m<sup>2</sup> (10 m x 10 m) was done by dividing the studied surface into 100 sub-plots (1 m x 1 m).

For each sub-plot we evaluated the area covered with invasive plants ( $\text{m}^2$ ). The data obtained in this way were analysed to obtain spatial distribution of species, and the coverage index for the studied species, which is the ratio of the total area covered by the target species (sum of the areas covered in 100 sub-plots) to the total area of the plot ( $100 \text{ m}^2$ ). Using these we calculated the spreading coefficient, which represents the increase in the area covered by invasive plant species starting from a reference area of  $1 \text{ m}^2$  during two years ( $\text{year}^{-1}$ ). Statistical methods used were Durbin-Watson test, and Fisher-Snedecor test where the dependent variable is the coverage index of the invasive species and the independent variables are: the air temperature, the rainfalls amount, the air relative humidity and the soil surface temperature.

## Results and discussion

The model of the surface occupation by the grassland invasive species from our research is radial, as the model described by Cousens and Mortimer (1995) for weeds. Thus the initial surface covered is increasing with a half from the radius of the preceding generation, and the expansion rate is constant. Small deviations from radial spread appeared in all cases analysed here, probably because some individuals from the initial population are spreading further from the invasion source forming new satellite populations (Maxwell *et al.*, 2003). The satellite populations function as new invasion sources continuing to increase as the original source of invasion. One of the analysed species did not follow these expansion patterns. This was *Xanthium spinosum* which, in 2004, was doubling the spreading coefficient from  $1 \text{ m}^2$  to  $2.08 \text{ m}^2$  manifesting an explosive increase of the population, and in 2005 the spreading coefficient was  $2.24 \text{ m}^2$ . An explanation for this could be the one formulated by Woodborn and Briese (1996), that the environment became improper for its own species through the accumulation of some chemicals that were disturbing the development of their own descendants. Thus, all the spreading coefficients calculated for the target species are greater than one; those are showing that the analysed invasive species are increasing the surface covered by them in a very short time. The statistical analysis shows for every model analysed here that the correlation coefficient value is greater than 0.8 (Table 1), suggesting the existence of a direct and moderate relationship among the analysed variables. The positive value of  $R$  for all these correlations shows that an increase of the independent factors (air temperature, rainfall amount, air relative humidity and soil surface temperature) is increasing the value of the dependent factor, the coverage index of the invasive species. The moderate relationship shows that there are other environmental factors that are influencing the coverage index of the invasive species too, but they were not included in this study. The same aspect is confirmed by the values of the  $R^2$  values, those values being greater than 0.6. The Durbin-Watson test (D-W) is used to verify the null hypothesis ( $H_0$ ) or the lack of autocorrelation of the residual values. The interval ranged between 0 and 4. The values obtained for D-W test for all three years are comprised between 2.269 and 2.45. Analysing the results obtained for the F test, these show relatively great values of  $F$  those being powerful arguments against to the  $H_0$ . The results obtained for the air temperature and for the soil temperature can be explained through the fact that most of the plants studied in this work are mesothermal or mesothermal – moderate thermophilic. Positive correlations between temperature increase and the habitats invasiveness have been obtained by Dukes and Mooney (1999), but they were referring to the increase of the incidence of the species invasion in ecosystems situated at great altitudes and latitudes. Similar results in this context were obtained by Pyšek (2001). Comparing the data concerning the grassland invasive plants needs for water, we have obtained data that show that the herbaceous species need less water in comparison with the shrub species (Sărățeanu, 2006). The species studied in this work are xeromesophytic and xeromesophytic-mesophytic from the point of view of their water needs.



Table 1. Statistical analysis of the influence of the independent factors (air temperature, rainfall amount, air relative humidity and soil surface temperature) on the invasive species coverage index during 2003-2005.

Specification	2003	2004	2005
Correlation coefficient ( <i>R</i> )	0.813	0.803	0.897
R Square ( <i>R</i> <sup>2</sup> )	0.661	0.644	0.893
Durbin – Watson test ( <i>D-W</i> )	2.450	2.269	2.336
Fisher-Snedecor test ( <i>F</i> )	3.463	3.845	134.667
Factor with positive influence	Rainfalls: 1.037 Air temperature: 0.271 Soil temperature: 0.160	Rainfalls: 0.121	Rainfalls: 0.124
Factor with negative influence	Air rel. humidity: -0.076	Air temperature: -0.418 Soil temperature: -0.051 Air rel. humidity: -0.665	Air temperature: -0.115 Soil temperature: -0.168 Air rel. humidity: -0.223

*F* = 2.84 for this study in case of *P* < 0.05.

## Conclusions

Based on the experimental results obtained we can conclude that the target species analysed from the point of spreading coefficient are acting as invasive species or weeds following a certain spreading pattern, except *Xanthium spinosum*. Analysing the influence of some climatic factors (air temperature, rainfalls amount, air relative humidity and soil surface temperature) on the coverage index of the invasive species there are positive correlations, but the dependent variable (coverage index) is influenced during this time by other factors too.

## Acknowledgements

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# Prediction of the potential effects of increased flooding periods on grassland in the core area of the German-Polish National Park

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## Abstract

For the amendment of the National Park law as enforced in 2006, it was necessary to be able to predict the potential effects of setting up a natural flooding regime (extending of the opening of the polder intake construction by one month) on the composition of the grassland plant communities, grassland yields and quality. The following could be deduced:

- the detailed limitation of the reserves I (total reservations) and II (ecologically compatible, extensive grassland utilization) 50:50% each;
- conflict fields with regard to protection of species with changes in the water regime;
- the long-term agricultural utilization of zone II and the yield situation to be expected there;
- a total summing up of the remaining usable agricultural areas, including yield and quality of fodder.

Keywords: overflowing, plant communities, yield, fodder quality

## Introduction

The centre zone of the national park on the German side comprises approximately 4,000 ha of grassland between Hohensaaten-Friedrichsthaler Wasserstraße (West Oder) and present-day Oderstrom (East Oder). This area will, in future, be subject to a natural flooding regime in the winter season and occasional summer flooding. The current flooding period is from 12 December until 15 April, and in the future period will be extended by one month.

## Materials and methods

The task could not have been solved without long-term and continuous monitoring preparation work. Sociological flora maps of the entire over-flooding areas (1:10,000) were drawn up in 1970 (Schalitz 1970), in 1997 (after the flood of the century), in 2002 (after regeneration of the crops) and in 2005 directly before drawing up the forecast.

Extensive ecological population measuring programmes were carried out on the important agricultural species (Schumann *et al.*, 1998; Schalitz *et al.*, 2002) to identify their reactions to environmental influences. Many years of yield and quality analyses were collected in a work brochure of Schalitz and Rogge (2002).

## Results and discussion

The over-flooding factor in the Oder floodplain led to a drastic reduction of biodiversity among grassland flora. It could be noted in general in the sociological flora area maps, that occurrence of prevailing plant communities depended on the height (relief formation) and, thus, on the duration of over-flooding and the ground water level.

The typical plant communities of the floodplain are:

a) *Alopecuretum pratensis* (reg. 25)

b) *Phalaridetum arundinacea* (Libb. 31)

Whereby meadow foxtails primarily continue to colonize the dry and cultivated areas

1 *Alopecuretum pratensis* (Reg. 25)

1.1 *Leontodon autumnal* sub-association (Autums dandylium-sub-community)

1.1.1 *Festuca pratensis*-Facies 1.1-2.2m a. NN  
(meadow fescue I – formation)

1.1.2 *Leontodon autumnale*-Facies 0.8-1.1m a. NN  
(Autumn dandylium-formation)

1.1.3 *Phalaris arundinacea*-Facies 0.8-1.0m a. NN  
(Canary grass-metamorphosed)

1.2 *Alopecurus geniculatus*-*Agrostis stolonifera*  
Sub association

(meadow fescue-creeping bentgrass sub-community)

1.2.1 *Alopecurus geniculatus*-*Agrostis stolonifera*-Facies 0.6-0.9m a. NN  
(meadow fescue-creeping bentgrass-formation)

1.2.2 *Phalaris arundinacea*-Facies 0.5-0.8m a. NN  
(Canary grass formation)

2 *Phalaridetum arundinacea* (Libb. 31) < 0.5m a. NN

In the population ecological measuring programme with *Alopecurus pratensis* in 1995-1998 at 6 plots of different elevations and, thus, flooding duration, it could be demonstrated how morphological features change in favour of the location, to the disfavour, respectively. After the flood of the century in 1997, the dry locations revealed clear advantages. If the flooding period is extended beyond the tolerance limits, the indicator plant *Alopecurus pratensis* dies back and, as a rule, a *Phalaridetum* or flood meadow develops (Figure 1). This is, as a rule, not desirable from the agricultural point of view (quality of fodder, diversity of usage).

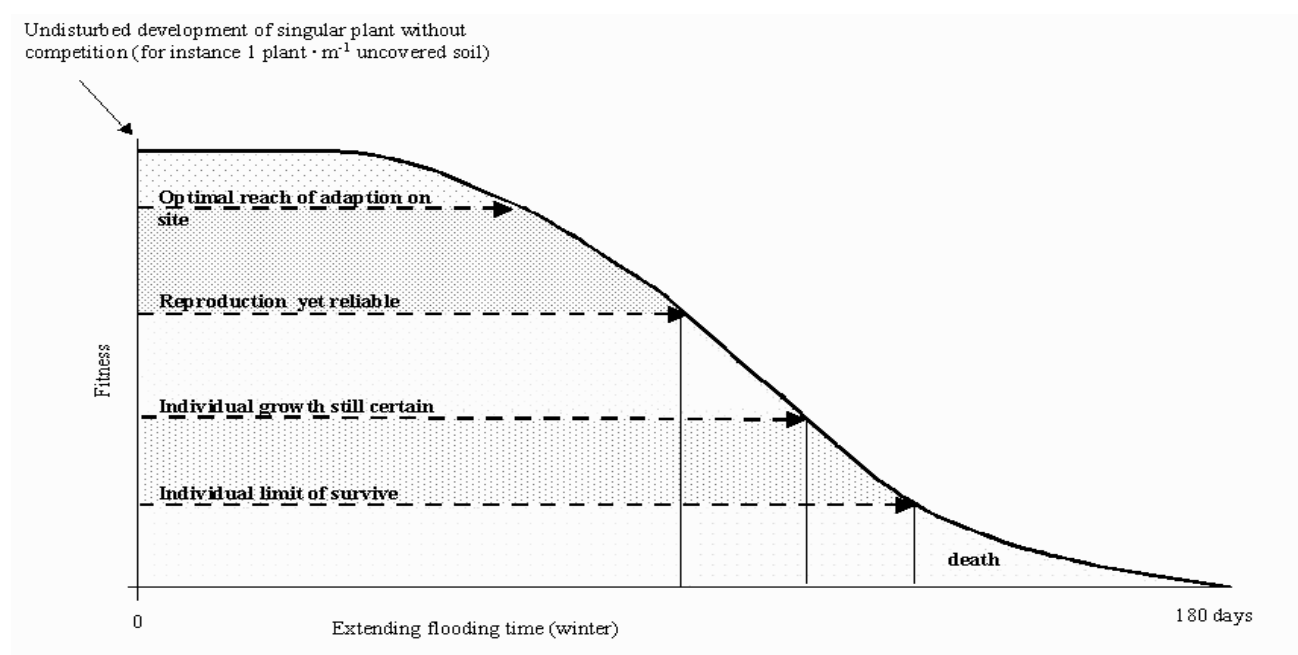


Figure 1. Connection between duration of flooding and the efficiency of meadow foxtail population.

To be expected when the flooding period is extended by 30 days:

- The wetland foxtail community will, to a great extent, shift to flood meadows. Thus, grasslands instead of meadows will develop (Protection Zone II).
- The dry to fresh foxtail community will metamorphose to the wetland form and remain good for grazing; however, the grazing period will begin later. We have valuable plants here now: *Trifolium repens*, *Trifolium hybridum*, *Vicia cracca* etc.
- There will be a shift to sedge communities under the influence of longer periods of flooding water (Classification primarily Protection Zone I).

The yields and fodder qualities of the individual plant communities will not change significantly, however the share of areas between the various farmers. A swap of land could be worked out based on the forecast map via the farmland reclassification process. Great losses in yield are to be expected by the users of wetlands. In wetzones of *Phalaridetum arundinacea*, *Carex* species spread massively, as well as herbs including *Senecio aquaticus*, *Rovippa amphibia*, *Ranunculus repens*, *Polygonum amphibium*, *Lythorum salicaria*, *Caltha palustris*, *Thalictrum flavum*, *Sium latifolium* and many others. The farmers are to be compensated with farmland located beyond the wetlands (Garzer Bruch).

The extended over-flooding period is not without consequences for nature conservation. Due to the change of utilization, other species, for instance *Cnidium dubium*, and reed and sedge warblers, will resettle.

## Conclusions

The fresh foxtail (*Alopecurus*) community will change to the wetland facies and remain good for pasture; however, the length of grazing period will be reduced. This high quality and productive foxtail forage is often entwined by leguminous tufted vetch (*Vicia cracca*). The wetland foxtail community will, to a great amount, shift to the *Phalaris arundinacea* community resp. *Phalaris arundinacea*-Facies of *Alopecuretum*. As a result of a shift in the wetland, a stock of sedge develops from the *Phalaridetum*, and this is not usable for agriculture.

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# Long-term effects of N, P and K fertilization on specific biodiversity in a permanent mountain meadow

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## Abstract

We examined the effects of N, P and K fertilization on specific biodiversity in a permanent mountain meadow located in the Dolomiti Bellunesi National Park (NE of Italy). Mineral fertilization significantly affected grassland productivity and specific biodiversity. NPK was the most productive treatment, and brought about a significant reduction of specific biodiversity. The highest number of species was recorded in the control. Although no increases in yield level were found, significantly lower values of specific biodiversity were detected in K and NP plots. Fertilizer addition caused a significant change in floristic composition compared with the control, but the rate of change in species composition differed among treatments. In PK plots, which indicated an increase of dry matter yield, no reduction of biodiversity was assessed. The increase of small-scale structural heterogeneity in the PK treatment represented a main factor in the maintenance of biodiversity.

Keywords: mineral fertilization, yield level, biodiversity, species succession

## Introduction

During the last 50 years mineral fertilizer application has been a common treatment for improving nutrient-poor grassland communities. The increase in the use of mineral fertilizers has frequently caused the loss of species biodiversity in oligotrophic grasslands (Schellberg *et al.*, 1999); consequently, the relationship between productivity and diversity has become a major issue in conservation management, and recently a great deal of effort has been dedicated to discovering the mechanisms that regulate species coexistence and succession. Along natural gradients of productivity, diversity often shows a unimodal pattern; however, considering nutrient enrichment experiments in grasslands, plant diversity generally decreases when fertilizer is added (Crawley *et al.*, 2005). Beside this, specific nutrients can have characteristic impacts on plant species richness. Therefore, a more detailed knowledge of the effects of soil nutrient supply on specific biodiversity and floristic succession is needed.

## Materials and methods

The present work was set up in a fertilization trial that started in 1977 and is still ongoing on a mountain permanent meadow (420 m a.s.l.) in the locality of Candaten (Sedico-Belluno, NE of Italy), within the Dolomiti Bellunesi National Park. An annual average temperature of 10.6 °C and a mean annual rainfall of 1,535 mm characterize the climate of the study area. The floristic composition of the meadow could be ascribed to the *Arrhenatherum elatioris* association, and to the *Salvietosum pratensis* sub-association; *Briza media*, *Festuca rupicola*, *Leontodon hispidus* and *Sedum sexangulare* were the most abundant species. In this work we analysed the following treatments: 0 (= control), N, P, K, NP, NK, PK and NPK (Table 1). The experimental design was a randomized complete block with 3 replicates. Plot size was 24.0 m<sup>2</sup>. In each plot 3 cuts have been taken each year. During 2004-2005 a botanical survey (Braun-Blanquet) was performed just before the first cut. Specific biodiversity (i.e. number of species) and cover value of all species were assessed in each treatment. Grassland yield at the

first cut was also measured by weighing plant biomass. In addition, using a 64 photodiodes ceptometer (Sunscan, DT Devices), per cent photosynthetically active radiation (PhAR) and spread values (i.e. the value of the standard deviation of the 64 photodiodes readings divided by their mean) at soil surface were measured. The rate of change in botanical composition of each treatment over the control was calculated using the Sorensen Similarity Index,  $SSI = 2a/b + c$ , where  $a$  represents the number of species in common between the treatment and the control,  $b$  is the number of species recorded in the control and  $c$  is the number of species observed in the treatment. Finally, for each treatment we measured the proportion of species in common with the control ( $SCC$ ), the proportion of new species (i.e. the proportion of species of the treatment that were not observed in the control,  $NSC$ ) and the fraction of species that were lost over the control (i.e. the proportion of species of the control that were not present in the treatment,  $SLC$ ).

## Results and discussion

Single fertilizer additions (N, P and K plots) did not lead to significant changes in grassland productivity (Table 1); with the only exception of NP plots, the improvement of grassland productivity depended on the combination of at least 2 fertilizers (NK, PK and NPK plots). A significant reduction of specific biodiversity was measured in the case of single K addition, but no changes were observed in the case of N and P plots. Concerning functional groups, N and K addition reduced grasses, while single P fertilization led to a decrease of legumes. Despite the increase in aboveground phytomass in binary and ternary treatments, only NPK plots denoted a significant reduction in total species number. The lowest values of biodiversity were observed in NP plots (-64.68%). These results suggested that the mechanisms responsible for the decline in species richness consisted of more than just increased biomass production. Similarly, PhAR values at soil surface (Table 2) could not directly explain changes in specific biodiversity, since the extremely low values detected in PK and NPK plots were not compensated for by a proportional decline in total species number.

Table 1. Fertilization rate, biomass yield and specific biodiversity values of the Candaten grassland. In each column, values that share no letters are different at  $P = 0.05$  Duncan's test.

Treat.	Fertilizer rate (kg ha <sup>-1</sup> y <sup>-1</sup> )			Yield (t ha <sup>-1</sup> )	Number of species			
	N	P	K		Total	Forbs	Grasses	Legumes
0	0	0	0	1.45 d	32 a	17.6 ab	11.6 a	2.8 ab
N	96			1.65 cd	26.6 ab	15.3 a-c	9.3 bc	2.0 b
P		108		1.54 d	31.0 a	18.8 a	11.5 a	0.6 c
K			216	1.47 d	23.3 b	13.0 bc	8.0 c	2.0 b
NP	192	108		1.38 d	11.3 c	5.8 d	5.3 d	0.2 d
NK	192		108	2.02 c	25.8 ab	14.1 a-c	9.1 c	2.5 ab
PK		54	216	2.63 b	26.6 ab	11.8 c	11.1 ab	3.7 a
NPK	192	54	216	3.31 a	22.5 b	11.5 c	8.5 c	2.5 ab

Fertilization caused a significant modification of floristic composition over the control, but the rate of change differed among treatments. In fact, the  $SSI$  values ranged between 0.58 and 0.64 in N, P, K, NK and PK plots, with clearly lower values in treatments NP (0.27) and NPK (0.39); only 50% of the plant species composing these treatments were also found in the control, indicating a clear differentiation of species composition with fertilization. In treatment NP, 81.2% of control species disappeared, with the loss of almost all legumes (-96.6%), tall forbs (-90.4%) and small forbs (-86.9%), while only small grasses persisted (*Anthoxanthum odoratum*, *Festuca rubra* and *Festuca rupicola*). New species were represented by small grasses, small forbs (as *Cerastium fontanum*) and tall forbs (*Silene vulgaris* and *Arabis hirsuta*). In contrast, K plots maintained most of the control species (78.4%), but



did not show a strong appearance of new species (21.6%); these were represented mostly by small forbs, such as *Carex montana*. In the most productive treatments *SLC* values were proportional to yield level; this tendency was also confirmed in each functional group. This occurred also for *NSC*, but single functional groups showed the highest values in PK plot. Treatment NPK denoted the decline of 61.1% of small grasses and 83.9% of small forbs; regarding tall forbs and tall grasses, a replacement of species occurred, with the spreading of some new productive species, such as *Arrhenatherum elatius* and *Trisetum flavescens*. Treatment PK showed a strong reduction of small unproductive forbs (*Sedum sexangulare*, *Carex caryophyllaea*); in addition, appearance of new species occurred among all functional groups. The high *spread* values observed in PK treatment suggest the presence of a mixture of areas with high and low light levels within the canopy: in this treatment the diversity of microhabitats could have been a key factor for the maintenance of many plant species characterized by different ecological requirements.

Table 2. Sorensen Similarity Index (*SSI*), *PhAR* values, *spread* values, per cent fraction of species in common with the control (*SCC*), per cent fraction of new species (*NSC*) and per cent fraction of species lost over the control (*SLC*) in the analysed treatments.

	N	P	K	NP	NK	PK	NPK
SSI	0.64	0.64	0.64	0.27	0.67	0.58	0.39
<i>PhAR</i> (%)	45.6	51.1	44.9	39.3	22.3	11.5	8.8
<i>Spread</i>	0.81	0.59	0.69	0.71	0.92	1.19	1.13
<i>SCC</i> (%)	71.2	65.5	78.4	52.8	73.6	65.5	50.8
<i>NSC</i> (%)	28.8	34.4	21.6	47.1	26.3	34.4	49.1
Tall forbs (%)	16.0	31.5	20.6	36.7	44.7	16.8	33.8
Small forbs (%)	62.5	31.4	57.7	31.1	27.0	28.8	29.1
Tall grasses (%)	9.0	22.5	10.3	2.3	20.0	22.2	18.2
Small grasses (%)	9.0	11.5	4.7	29.6	2.0	19.9	10.7
Legumes (%)	3.3	3.0	6.5	0.0	6.1	12.0	8.0
<i>SLC</i> (%)	40.1	34.3	43.0	81.2	39.3	45.0	65.9
Tall forbs (%)	49.3	13.2	41.3	90.4	35.3	37.2	67.5
Small forbs (%)	41.1	42.9	53.3	86.9	57.6	75.7	83.9
Tall grasses (%)	45.7	33.6	42.0	86.2	31.2	35.4	50.5
Small grasses (%)	5.5	26.3	31.9	29.1	34.7	34.7	61.1
Legumes (%)	40.0	88.3	40.0	96.6	18.3	15.0	47.5

## Conclusions

In Candaten grassland specific biodiversity was not clearly correlated to sward productivity and *PhAR* level at soil surface. In spite of a different behaviour of single functional groups, a significant reduction of total species richness was recorded only in treatments K, NP and NPK. In plots where a significant increase of aboveground yield occurred, botanical composition changed toward a more competitive community, with the appearance of new species distributed among all functional groups. In PK plots, the high heterogeneity in light availability could have represented a main factor in the maintenance of specific biodiversity.

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# Botanical and ecological description of pasture types in an area of the Italian Alps

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## Abstract

Alpine landscape is the outcome of the interactions between environment, vegetation and local human activities. Knowledge of the vegetation types present in mountain areas is an important tool for the proper management of these environments, which are delicate and threatened by the socio-economic changes of recent decades. The aim of this study was to create a handbook for the description of the most important pastoral types present in the area. The study was carried out in Comelico (Italian Alps) on a pastoral surface area of > 2,000 ha. The research is based on about 350 botanical analyses, on the application of multivariate statistics, and on the Landolt ecological indexes for the discrimination of different identified types. The survey permitted the identification of six main pasture types, and it allowed both ecological and botanical description of the different pasture communities in order to deepen the knowledge on the characteristics and on the dynamics of the Comelico pastures.

Keywords: pasture types, management, Landolt indexes, multivariate statistics

## Introduction

The development of industry and tourism, and rural depopulation in recent decades, are the key causes in the decline of agricultural practices throughout the Alps. Alpine grasslands are undergoing a reduction in surface area (through forest recovery) and a floristic alteration (through shrub encroachment) due mainly to stocking rate reduction, irrational management and poor knowledge of pastoral ecosystems (Gusmeroli, 2004). Many efforts have been made on several Alpine territories with the aim of pasture characterization. The study reported here was carried out in Comelico (eastern Italian Alps), an inner Alpine territory where the decline in agriculture has been particularly severe, with 52% reduction in meadow and pasture surface and 74% reduction in livestock between 1970 and 2000. The main object of the survey was the development of a pastoral typology, which is a pasture characterization tool able to improve the knowledge about ecology, management and production of grassland areas.

## Materials and methods

The study was carried out in the eastern Italian Alps (Carnic Alps) on the border between Italy and Austria on 2,129 ha of pasture surface. The botanical characterization of natural upper-level pastures was surveyed in three contiguous valleys: Digòn Valley, Visdende Valley and Sesis Valley. The research is based on about 350 point quadrat transects following the phyto-pastoral method (Daget and Poissonet, 1969) for the botanical characterization and pastoral value assessment which considers both forage quality and production. Elevation, exposure and slope were also recorded. After a data reduction for redundancy diminution, the species-specific contributions were ordered by cluster analysis (Bray-Curtis distance, Ward agglomeration method). A fuzzy clustering, which provides the membership percentage of each survey to each type (Kaufman and Rousseeuw, 1990), was applied to the data to obtain a deeper knowledge on the classification structure. Landolt ecological indexes (Landolt, 1977) were applied to the botanical data in order to obtain an indirect ecologic characterization. The ecological values obtained multiplying species-specific contribution in each survey by

Landolt ecological indexes (F humidity, R pH reaction, N nutritive elements, H humus, D substratum coherence, L light, T temperature, K continentality) and the environmental parameters (slope, elevation, distance from shelter) collected during the surveys have been used for the pastoral types description and characterization through correlation analysis.

## Results

The interpretation of the cluster diagram made possible the definition of 6 pastoral types in the study area. The ordination was based on the species composition: type 1 dominated by *Carex curvula* and *Nardus stricta*; type 2 dominated by *Nardus stricta*; type 3 dominated by *Rhododendron ferrugineum* and *Vaccinium myrtillus*; type 4 dominated by *Sesleria varia* and *Carex sempervirens*; type 5 dominated by *Festuca* gr. *rubra*; type 6 dominated by *Deschampsia caespitosa*. The types are homogeneous from the point of view of botanical composition and, employing the same ordination diagram, it has been possible to split the types into 15 ecofacies which are homogeneous from the management point of view, and therefore they represent the main pastoral unities. Type 1 was the most widespread with 676 ha total surface; by contrast the least common was type 4 with 131 ha. The classification obtained with the cluster analysis was confirmed by the application of a fuzzy clustering. The surveys classified by cluster analysis have a clear membership percentage with the corresponding type (Table 1); only types 1 and 5 reached a membership coefficient below 50% (49% and 45% respectively), while types 2, 3 and 6 were clearly differentiated from the others with more than 60% on average of membership coefficient. The main overlappings are between type 2 and types 1 (42%), 3 (21%) and 5 (33%), which could indicate the central position of type 2 in the studied pastures transitions.

Table 1. Comparison between hierarchical and fuzzy clustering, types membership and correctly classified (all data %).

	Type 1	Type 2	Type 3	Type 4	Type 5	Type 6
Type 1	49	12	8	4	5	3
Type 2	42	68	21	19	33	18
Type 3	5	4	65	3	2	1
Type 4	0	1	1	52	5	1
Type 5	3	13	4	12	45	14
Type 6	1	2	1	10	10	63
Correctly classified	49	68	65	52	45	63
Fuzzy	51	32	35	48	55	37

The correlation between type membership percentages and ecological and pastoral values, and environmental parameters, could give important information on the most important factors in pastoral vegetation differentiation (Table 2). F index results are the least significant in vegetation types discrimination, while R and N indexes are highly significant because they separate acid and poor fertility types (1 and 3) from more neutral and rich types (5 and 6). In particular, R and D indexes confirm the localization of type 4 in calcareous-rich areas characterized by a more elevated pH and a more incoherent substratum. L index underlines the difference between 'open' vegetation types (1, 2 and 5) from type 3 dominated by shrubs which favour shade-tolerant species. Furthermore, type 3 is characterized by high H values because of the litter accumulation under the shrub formations. Continuous grazing is the main management technique in the area; animals tend to avoid sloping areas where shrub species are becoming dominant: this is confirmed by the correlation between sloping areas and shrub vegetation (type 3). Elevation and distance from shelter are useful parameters in the discrimination between type 1, linked with upper level areas, and type 5, common in the neighbourhood of shelters which are situated in the lower pasture areas. N index and pastoral

value have a tight relationship, which indicate a higher forage quality in the rich areas. Types 1, 3 and 4 are the less important from the productive point of view, whereas type 2, and overall, type 5 are the most productive. Type 6, though having high N index values, has a low pastoral value because it is characterized by the presence of nitrophilous flora.

Table 2. Pearson correlation coefficients between pastoral value, ecological indexes and environmental parameters following fuzzy analysis types membership (univariate test, \* =  $P < 0.05$ ; \*\* =  $P < 0.01$ ).

	Type 1	Type 2	Type 3	Type 4	Type 5	Type 6
Pastoral value	-0.124*	0.168**	-0.264**	-0.138**	0.392**	-0.016
<i>Ecological indexes</i>						
F	0.007	0.040	-0.031	-0.025	-0.064	0.069
R	-0.214**	-0.025	-0.342**	0.231**	0.303**	0.159**
N	-0.141**	0.116*	-0.282**	-0.067	0.246**	0.162**
H	-0.065	-0.061	0.328**	0.048	-0.174**	-0.116*
D	-0.064	0.091	-0.040	-0.207**	0.155**	0.034
L	0.163**	0.209**	-0.354**	-0.059	0.123*	-0.054
T	-0.342**	0.040	-0.041	0.038	0.260**	0.072
K	-0.048	0.124*	-0.149**	-0.086	0.157**	0.010
<i>Environmental parameters</i>						
Slope (%)	-0.070	-0.096	0.147**	0.070	-0.086	0.038
Elevation (m a.s.l.)	0.410**	0.131*	0.071	-0.039	-0.546**	-0.083
Distance from shelter (m)	0.159**	0.089	-0.001	0.086	-0.251**	-0.086

## Conclusions

Multivariate statistical analysis techniques proved useful in the identification of vegetation groups, and the interpretation of results allowed the characterization of six pastoral types in the studied area. The fuzzy clustering confirmed the classification obtained by hierarchical analysis and it raised the information level about relationships between vegetation types. Correlation between membership types and ecological indexes was useful in the characterization of types of different ecological spaces; in particular R and N indexes had high statistical significance. Environmental factors had a lower significance but they indicated a narrow relationship between shrub vegetation and sloping areas and they were useful in the discrimination between types 1 and 5. Pastoral value results were significant factors in the differentiation between pastoral types. Pastoral types represent a useful tool as they allow a targeted extensive utilization considering the different pastoral surfaces and, therefore, facilitation for rational management.

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# Feral sheep in coastal heaths – developing sustainable agriculture in vulnerable cultural landscapes

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## Abstract

This is an interdisciplinary project (2007-2010) where researchers on landscape ecology, animal health, and agricultural economy join forces in order to meet the main objective; develop feral sheep farming into sustainable agriculture for the 21st century. The project is based on the idea that a combination of innovation and management will be the most efficient and cost-effective tool for the future national care of coastal heathlands. This unique semi-natural landscape is managed by year-round grazing by feral sheep, an old domestic breed, in combination with heath burning. The heathland has a complex successional dynamic in which the right balance between winter and summer fodder is of vital importance for its sustainability. As a result of land-use change and abandonment, the coastal heath ecosystem today is classified as 'greatly endangered', and the grazing quality of several heath areas are reduced. Some farmers have difficulties securing carcass weights of feral sheep. This might be caused by several confounding factors. In this project cobalt deficiency is shown to be the main explanation at one locality. However, at other localities the answers may not be so straightforward.

Keywords: vulnerable landscape, coastal heathlands, feral sheep

## Introduction

Year-round sheep farming on the Norwegian west coast has a history dating back several millennia. This important part of our cultural heritage has resulted in a distinct breed of animals, the feral sheep. This breed has evolved in close interaction with the shaping and maintenance of a unique semi-natural landscape: the coastal heathlands. Heathlands are usually confined to places with a humid oceanic climate with mild winters, which gives an opportunity for year-round grazing. Sheep have been the most common domestic animals on heathlands. Burning regimes are necessary for maintenance of heathland ecosystem cycles and creation of a mosaic of grasslands and heather for year-round free-range grazing. Feral sheep prefer the more nutrient-rich grasses and herbs during spring, summer and autumn, but depend on a sufficient supply of evergreen plants, especially heather, during winter (Clarke *et al.*, 1995; Kaland, 1999). However, the natural conditions along the Norwegian western coast suggest that feral sheep may be exposed to insufficient supplies of essential minerals and trace elements. Both the bedrock and the superficial deposits are generally acidic and poor in mineral nutrients. Furthermore, deposits of shell sand with high pH levels can inhibit the uptake of certain trace elements in plants (Whitehead, 2000), and the high precipitation in oceanic regions can further reduce availability of minerals and trace elements (Låg, 1968; Steinnes, 1988). Detailed knowledge of how to manage these heterogeneous landscapes to secure sufficient amounts and qualities of different fodder plants is, therefore, of crucial importance for animal health, growth, and well-being.

## Materials and methods

This interdisciplinary project (2007-2010) deals with ecological, economic and animal health studies. Among other things, weights and blood samples of lambs of feral sheep have been collected at eight localities in order to assess the animals' well-being, particularly the prevalence of cobalt deficiencies. The study sites are from three different regions along the western coast of Norway: one site in Lindås (Hordaland), two sites in Solund (Sogn og Fjordane) and five sites at Frøya (Sør-Trøndelag). All sites are situated in areas where cobalt deficiency can be suspected. The lambs at the study sites have been divided in two balanced groups of both genders. (For more details see Figure 1 and Table 1). In June one of the groups was treated with a supplement of cobalt, the other group remained as a control. Weights are taken in June (at the same time as treatments are given) and October. In addition the farmers will provide the carcass weight of the lambs (all male, most females). Furthermore, a more detailed study has been carried out at another locality at Lindås (Lindås 2, close to Lindås 1; see Figure 1 and Table 1). At Lindås 2 the weights and blood samples have been collected in June, August and October. Data from 2003 are presented in Table 1. The blood samples have been analysed for phosphorus, calcium, magnesium, copper, zinc, vitamin B<sub>12</sub> (cobalt), folate, liver enzymes ( $\gamma$  GT, GLDH), urea and total protein. All samples have been examined according to standard methods already applied in the laboratory of the Norwegian School for Veterinary Science, Section for Small Ruminant Research, Sandnes, Rogaland. Data from 2003 have been tested by two-sample t-tests. Preliminary data from 2007 have not yet been tested.

## Results

In 2007, Frøya 1 is the locality with lowest mean weights (Figure 1).

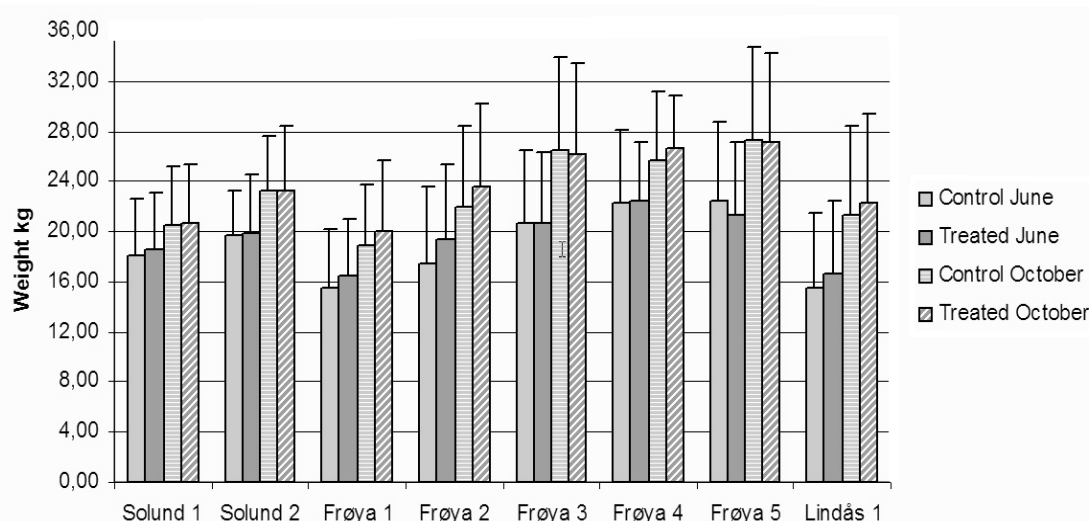


Figure 1. Mean weights (and standard deviation) of lambs, June and October 2007, at the different study sites (N in June and October = 26 and 26 at Solund 1; 24 and 24 at Solund 2; 22 and 14 at Frøya 1; 12 and 14 at Frøya 2; 28 and 28 at Frøya 3; 36 and 36 at Frøya 4; 24 and 24 at Frøya 5; 30 and 26 at Lindås 1).

At Lindås 2 (Table 1) the lambs treated with cobalt had significantly improved weights in August and October ( $P < 0.05$ ) compared with the untreated lambs (control). Here untreated lambs lost weight from August to October ( $-0.6$  kg), while the treated lambs gained weight



during the same period (+1.2 kg). Blood analyses showed that 5% of lambs from Lindås 2 had B<sub>12</sub> values less than 150 pmol l<sup>-1</sup> in June. In August and October this number rose to 85% and 100%, respectively, of the untreated lambs. This means that in October all untreated lambs had values under the limit associated with critical cobalt deficiency (150 pmol l<sup>-1</sup>, Ulvund, 1990). None of the treated lambs had values less than 150 pmol l<sup>-1</sup>.

Table 1. Mean weights and standard deviation for lambs in June, August and October 2003 at Lindås 2. The numbers of lambs in the groups are in parenthesis.

	June		August		October	
	Treated	Control	Treated	Control	Treated	Control
Lindås 2	12,9/2,0 (22)	13,3/2,1 (22)	23,6/3,7 (22)	20,7/4,2 (22)	24,8/3,3 (22)	20,1/4,4 (22)

## Discussion and conclusions

Figure 1 shows that there are some differences in lamb weights between the study sites. Frøya 3-5 and Solund 2 have weights that will provide required carcass weights (9-16 kg). The weights at Frøya 1-2, Solund 1 and Lindås 1, however, might provide carcass weights under the requirement (9 kg). Also at Lindås 2 the untreated lambs have low weights but the cobalt treatment influences the values of B<sub>12</sub> and results in adequate weights. However, as seen in Figure 1, low weights are not necessarily influenced by cobalt deficiency, i.e. the cobalt treatment does not always result in improvement of lamb weights. Several confounding factors might influence the weights and health of feral sheep. For instance, Frøya 3-5 have the largest amount of grassland compared with the other sites, and are also the sites with the best weights. Furthermore, sheep at Frøya 5 graze in a well-managed landscape with heather of better condition compared with the other localities. In this project we therefore want to investigate the importance of different factors, like bedrocks and deposits, the vegetation composition and quality of grazed plant species. The plant and vegetation preferences, and how the sheep exploit the heathlands throughout the year will also be studied. The coastal heathlands are relatively low-production semi-natural habitats. The way the sheep exploit the energy and protein during wintertime is of special interest. Heather is seen as the most important winter fodder but *in-vitro* analysis of heather (Ulvesli and Nordbø, 1945; Velle *et al.*, 2005) indicates that it has a low digestibility. In order to meet the challenges that face the feral sheep farming and to take care of the heathlands, we believe this interdisciplinary approach is necessary.

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# The botanical changes of the *Lolio-Cynosuretum* under different management conditions

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## Abstract

In the locality of Chvojníčka in the Strážov mountains (middle Slovakia) the botanical changes of the *Lolio-Cynosuretum* R. Tx. 1937 after the end of different utilizations were observed. The trial was established in 1986, in a block design with four replications, and was utilized up to 2002. After 2002 two management systems were applied: (1) without cutting (abandoned); (2) cutting once a year, leaving plant material after mulching. Botanical composition and its changes in 2003-2006 differed according to exploitation in the years 1986-2002. Grass cover decreased in both management systems but was more pronounced with mulching. Legumes and bare patches in the cover were found to be decreasing, whereas the herbs were increasing.

Keywords: end of utilization, mulching, abandonment

## Introduction

In Slovakia, livestock numbers have decreased since 1989 to one-third of the numbers formerly present (Green Report, 2007). The area of grassland utilisation has also decreased, and inaccessible areas have been abandoned, degraded and colonized by weeds.

The impact of cutting and grazing has been studied by many researchers in Slovakia (Jančovič, 1998) but mulching only to a small extent. Research on this topic has been carried out in the Czech Republic (Kvítek *et al.*, 1998). The aim here was to study the botanical changes after different management regimes of previous grassland exploitation.

## Methods

Experimental investigations were carried out in a long-term grassland trial. In the years 1986–1993, the effects of different fertilization intensity were studied. In 1994–1996 no fertilization took place and the vegetation was cut once a year with the aim of maximum above-ground phytomass production (Rychnovská *et al.*, 1987). In the period 1997–2002, the same fertilization regimes were performed as in the years 1986–1993. In this paper the results of the investigations during 2003–2006 are given. In 2003 the experimental plots were divided into two equal parts. One part was left abandoned, and in the second one the vegetation was mulched after cutting with the aim of maximum above-ground phytomass production (Rychnovská *et al.*, 1987).

In the years 1986–1993 the fertilizer regimes were:

1. Unfertilized control
2. PK – constant rate of phosphorus and potassium (35 kg P ha<sup>-1</sup> and 70 kg K ha<sup>-1</sup>), were applied in spring
3. PK + N<sub>60</sub> – 60 kg N ha<sup>-1</sup> was applied in spring
4. PK + N<sub>120</sub> – 80 kg N ha<sup>-1</sup> was applied in early spring, 40 kg N ha<sup>-1</sup> after the 1st cut
5. PK + N<sub>240</sub> – 100 kg N ha<sup>-1</sup> was applied in early spring, 80 kg N ha<sup>-1</sup> after the 1st cut and 60 kg N ha<sup>-1</sup> after the 2nd cut

The site is situated at an altitude of 640 m a.s.l. in the locality of Chvojnica in the mountain of The Strážov Hills (48°53'N, 18°33'E). It belongs climatically to a mild temperate region, medium dry with prevailing cold winter. According to long-term measurements, the average yearly temperature is 7.5 °C, and during growing season 11.1 °C. Long-term average annual total precipitation is 848 mm, and in the growing season 431 mm. The soil-forming substrate is derived from crystalline rocks, with a predominance of granite and crystalline slates, on which a brown, acidic, sandy-loam soil (Cambisol) has developed. In phytocenological terms the original grassland was represented by the *Lolio-Cynosuretum* Tx. 1937 association before the experiment was established. From the viewpoint of botanical species occurrence, grass species (73%) predominated in the grassland, leguminous plants formed 2% of the cover and other meadow herbs represented 25%. Dominance of the botanical groups and individual species were determined before mulching for areas under both managements, according to Regal (1956).

## Results and discussion

Before the management change (1986–2002) the botanical composition in the examined grassland was typical for the fertilization regimes. Detailed results from this period were published by Vozar (2003). Botanical composition from the investigated period (2003–2006) is given in Table 1.

Table 1. Botanical composition in the years 2003–2006 (%).

Botanical groups	Year	Variant									
		1		2		3		4		5	
		A	M	A	M	A	M	A	M	A	M
Grasses	2002*	23	52	40	34	62	69	52	69	79	81
	2003	22	63	34	46	47	47	46	65	79	83
	2004	13	61	36	40	48	41	46	56	73	83
	2005	9	54	27	54	33	36	46	50	71	73
	2006	44	60	19	60	44	44	40	42	76	72
	s <sup>2</sup>	183.7	22.5	68.7	109.2	107.7	162.3	18.0	120.3	12.8	29.8
Legumes	2002*	4	+	15	15	1	1	+	+	-	+
	2003	5	1	27	20	8	16	10	5	-	-
	2004	6	+	28	24	6	20	8	8	-	+
	2005	7	-	30	10	10	15	10	8	1	1
	2006	8	1	5	10	8	17	8	10	3	1
	s <sup>2</sup>	2.5	0.3	114.5	38.2	11.8	54.7	17.2	15.2	1.7	0.3
Herbs	2002*	45	20	27	11	27	25	38	27	16	17
	2003	43	24	37	14	33	34	40	27	13	16
	2004	47	29	35	18	37	37	43	32	17	15
	2005	55	26	45	28	45	48	39	39	18	24
	2006	28	39	66	30	48	39	37	40	14	25
	s <sup>2</sup>	96.8	51.3	221.0	71.2	74.0	69.3	5.3	39.5	4.3	22.3
Blank places	2002*	28	28	18	40	10	5	10	4	5	2
	2003	30	12	2	20	12	3	4	3	8	1
	2004	34	10	1	18	9	2	3	4	10	2
	2005	29	20	+	8	12	1	5	3	10	2
	2006	20	+	10	-	+	+	15	8	8	2
	s <sup>2</sup>	26.2	112.0	59.2	227.2	24.8	3.7	25.3	4.3	4.2	0.2

s<sup>2</sup> – variance, A – abandoned, M – mulching, + - rarely, - not present.

\*For better understanding of the botanical changes, the situation in 2002 is added.

The botanical composition was different in the year 2002, preceding the beginning of the investigation, due to the management regime which had been applied earlier. In general, higher rate of nitrogen, increased fraction of grasses and PK-fertilizing, supported the cover

of legumes. After the grassland management changed at the beginning of 2003, the botanical composition changed continuously during the whole evaluation period 2003-2006. Different effects of abandoned and mulched grassland on the botanical composition were shown, depending on previous management.

In fertilizer regime 1 A (abandoned) a decline of the grass fraction to 9% was observed during 2003-2005. However, the grass fraction increased to 44% in 2006. This increase of the grass fraction was accompanied by reduction of the herbs. Under the mulching treatment within the same regime (1 M) the fraction of grasses in the cover slightly increased but the presence of herbs increased nearly two times. It was a positive finding that there were no bare spaces in the grassland at the end of the evaluation period.

The most dynamic development was shown in regime 2. As in regime 1, different effects of abandoned and mulched treatment on the botanical composition were shown. The herbs increased in abandoned grassland and the bare spaces declined from 40% to 0%, which was particularly interesting.

In management regime 3, a decrease of the grass fraction was observed, especially in the first year after the change of management regime. This decline, as well as a reduction in the area of bare spaces, was compensated by increased presence of herbs at the end of the investigation period.

The most stable grasslands were those with the highest fertilization levels (regime 4 and 5), and they gave the same results for both management practices since the year 2002. More noticeable, a decline of the grass fraction was found in regime 4.

The decrease of bare spaces can be evaluated as a positive outcome. Our results are in accordance with Kvítek *et al.* (1998), who found that multiple mulching led to gradual increase of legumes in the cover, whereas in abandoned treatments the presence of herbs increased.

## Conclusions

In general, there was a tendency for decreasing proportions of grasses and increasing proportions of herbs in the grassland during the period of 2003-2006. The proportion of legumes increased in the majority of regimes under both managements. Some considerable changes in the botanical groups point to the need for future detailed study of population structures.

## Acknowledgements

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# Photoperiodic effects on elongation growth of two timothy ecotypes

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## Abstract

Seedlings of two latitudinal ecotypes of *Phleum pratense* L., 'Engmo' (69°N) and 'Grindstad' (59.5°N), were grown under six photoperiods, equidistant from 9 to 24 h. Leaf tip heights were recorded every other day. Other morphological traits were measured at harvests five times during the 70-day period. Plant reactions were described by a functional relationship to sequential leaf number and a photoperiodic response function including photoperiod (PP) and PP of maximum sensitivity (PP<sub>c</sub>). Height growth was strongly dependent on photoperiod, which influenced cultivars differently. 'Grindstad' responded mainly within a narrow PP interval, with PP<sub>c</sub> of 12.5 h. 'Engmo', with a PP<sub>c</sub> of 15 h, responded nearly linearly over most of the PP range.

Keywords: latitudinal ecotypes, leaf growth, phase change, *Phleum pratense*

## Introduction

Short-term leys sown with two or more of the best suited perennial grass species, and often also with red clover as a main component, constitute the major part of the intensively managed grassland of Norway. Timothy (*Phleum pratense* L.) has been, and still remains, the foremost ley grass species. The longevity of an individual component of a mixed plant stand is related to climatic, edaphic, and management factors. In perennial plants photoperiod is a prominent climatic factor affecting their seasonal growth rhythm, and thus their agronomic performance. However, for modelling purposes, functional relationships should be quantified. Thus, it was found highly relevant to investigate the photoperiodic effect on morphological traits of latitudinal ecotypes over an adequate range of photoperiods.

## Materials and methods

Seedlings of two Norwegian latitudinally adapted cultivars of timothy, 'Engmo' (69°N) and 'Grindstad' (59.5°N), were grown in pots for 70 days in growth chambers under six equally spaced photoperiods from 9 to 24 h. The treatments were obtained through daylength extension using incandescent lamps irradiating 1.5-1.6  $\mu\text{mol m}^{-2}\text{s}^{-1}$  for a 9 h photosynthetic light period with 155-170  $\mu\text{mol m}^{-2}\text{s}^{-1}$  irradiance. Air temperatures in the chambers were 16 °C, and 8 °C, respectively, during the 9 h photosynthetic period and the rest of the diurnal cycle. The tip heights of (vertically erected) elongating main tiller leaves above soil surface (LTH) were observed every other day. Harvests of 2 pots per cultivar and treatment were conducted after 35, 49, 62, and 70 days to record other relevant morphological traits. The asymptotic approximation of the individual LTHs to their ultimate heights (H<sub>max</sub>) during the days of elongation growth (t) was described by the logistic function:

$$LTH = H_{\max}/(1 + \beta e^{\gamma t}) \quad (1)$$

H<sub>max</sub>,  $\beta$ , and  $\gamma$  (in turn Y<sub>i</sub>) were all, with their own sets of coefficients, related to leaf sequential number along the axis (LN) and photoperiod (PP) (Wu *et al.*, 2004):

$$Y_i = Y_0 + aLN + bLN^a[e^{c(PP-PP_c)}/(1 + e^{(c+d)(PP-PP_c)})] \quad (2)$$

where  $Y_0$  is the regression constant,  $c$  and  $d$  are sensitivity parameters,  $PP_c$  is the photoperiod of maximum sensitivity, and  $\alpha$  accounts partly for the interaction between LN and PP.

## Results

Figure 1 shows various responses to photoperiod of main tiller leaf-tip heights, as predicted from the equations (1 and 2), yielding estimates well correlated to the bi-daily measurements ( $r^2 = 0.97$  for 'Engmo' and  $r^2 = 0.99$  for 'Grindstad'). Under short daylengths ( $\leq 12$  h) the leaf-tip height decreased with leaf number and thus with time from start of treatment, whereas at long day conditions ( $\geq 15$  h) leaf-tip height increased with sequential leaf number, except for 'Engmo' at 15 h.

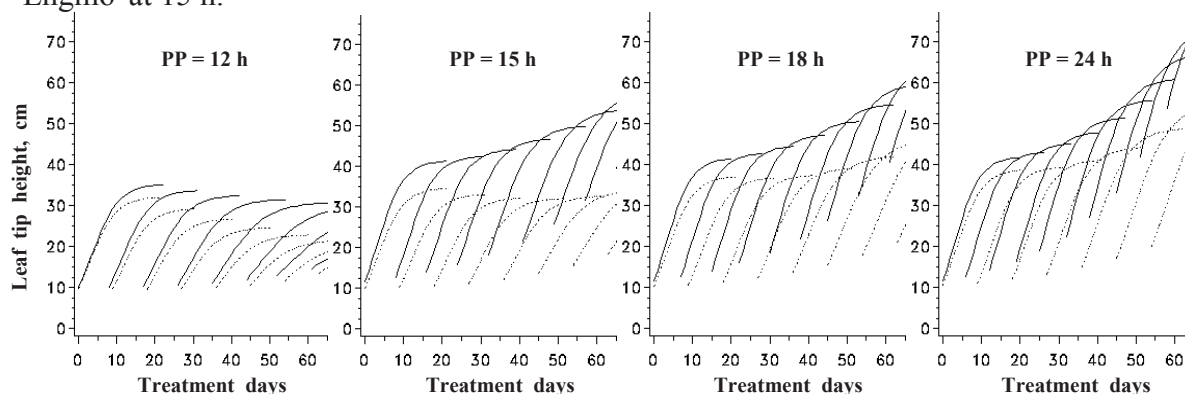


Figure 1. Predicted response to photoperiod in main tiller leaf tip heights from appearance to full elongation of leaf no. 6 and onwards in two timothy cultivars, 'Engmo' (--) and 'Grindstad' (—).

The leaves of 'Grindstad' grew longer than those of 'Engmo' under all daylengths throughout, but the relative difference was greatest just below 15 h, best visualized when expressing the above results as normalized response curves (Figure 2). 'Grindstad' revealed a steep slope located around 12-13 h, and had reached close to maximum ( $> 0.9$ ) at 15 h, whereas 'Engmo' showed a slower, almost linear reaction from 9 h to reach the same level (0.9) at about 20 h daylength.

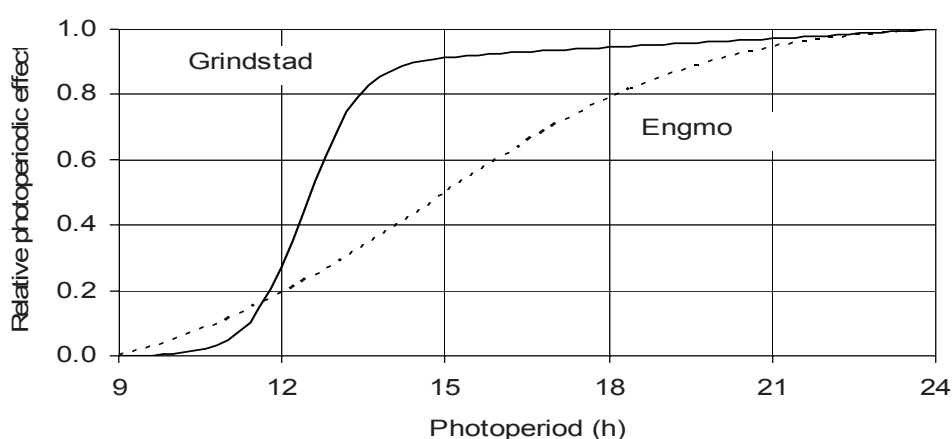


Figure 2. Normalized photoperiodic effect between 9 and 24 hours on leaf elongation growth expressed by leaf tip height above soil surface in two timothy ecotypes.

For daylength  $\geq 18$  h the differences between cultivars in height growth of leaf tips (Figure 2) were related to differences in stem elongation (Figure 3), which usually follow transition to

generative development. Any appreciable stem elongation occurred under daylengths  $\geq 15$  h in cv. 'Grindstad', and  $\geq 21$  h in 'Engmo', first by day 70. In 'Grindstad', stem elongation was observable by day 49 under 15 h daylength, proceeding steadily to reach some 32 cm at last harvest on day 70. By 21 h daylength, maximum length, *ca.* 45 cm, was reached in 62 days.

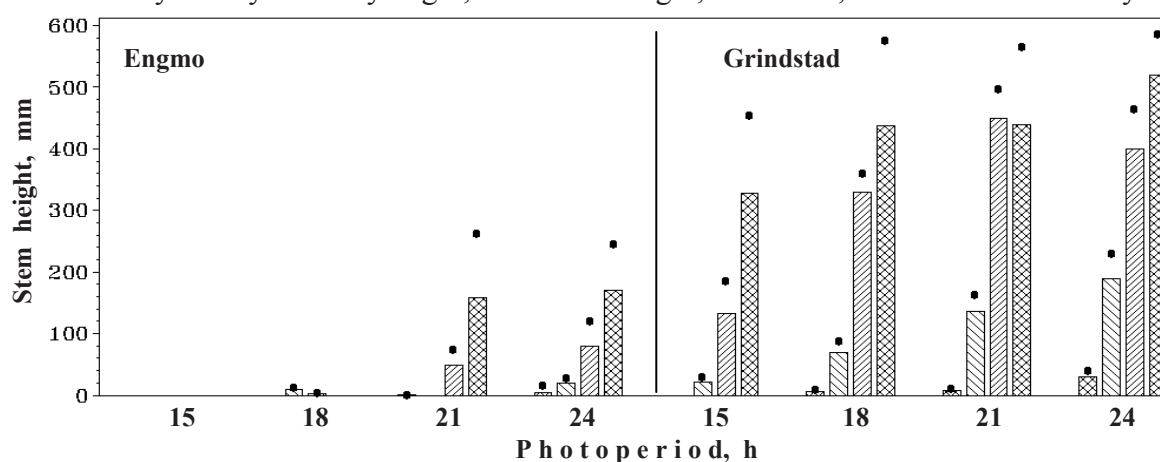


Figure 3. Main stem elongation growth (bars) plus one standard error of means ( $n = 4$ ) ( $\bullet$ ) as affected by photoperiod in two timothy cultivars grown at 11 °C (mean diurnal temperature) from the 6-leaf stage to each of four harvest days. The bars in each photoperiod group represent the harvest day numbers, from left: 35, 49, 62, and 70.

## Discussion and conclusions

Timothy is considered as the multipurpose grass species best suited for Norwegian conditions, due to its high productivity, quality and excellent winter hardiness. During its relatively long cultivation history in Norway this species has changed both in growth habit and winter hardiness due to adaptation to the growth and wintering condition of different regions. Thus, the cultivar 'Engmo', developed at 69 °N, has gained an excellent winter hardiness, seemingly to the cost of productivity, especially elongation and height growth, in late summer and autumn. Therefore, investigation of the close relationship between growth rhythm and longevity, and the effect of daylength in this respect, has been a main concern in the studies of the very nature of survival in marginal grass cultivation areas. This investigation reveals some of the strategies of a winter-hardy variety under short photoperiods, i.e. under late summer and autumn conditions: restricted growth, especially elongation and height growth – a part of which is exposed to loss by mowing or grazing – for the benefit of storing reserves in the leaf and stem bases.

The experiment demonstrates how a key external factor differentiates between ecotypes, and how a common functional relationship may account for very different responses to photoperiod. In modelling this is essential when relating contrasting growth habits to causal factors. Thus, the present quantification of one such relationship may be valuable in future grassland modelling, and we think that further experiments to clarify other key relationships will be worthwhile.

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# Floristic diversity and bioindicative assessment of the meadow habitats in the Huczwa river valley

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## Abstract

The permanent grasslands under study are situated in south-eastern Poland in the Lublin Province. The objectives of the research were the bioindicative assessment of meadow habitats in the Huczwa river valley and the determination of their floristic biodiversity. The investigation was based on 79 phytosociological records (relevés) taken using the Braun-Blanquet method, on meadows and pastures in the Huczwa valley, the property of private farmers. On the basis of the relevés the following were distinguished: 9 plant communities of the *Phragmitetea* class, 2 communities and 1 associations of the *Molinio-Arrhenatheretea* class and one community of the *Scheuchzerio-Caricetea nigrae* class. The *Caricetum lasiocarpae* community proved to be floristically richest, whereas *Phragmitetum australis* was the poorest. The meadow plants used as bioindicators showed that the phytocenoses occurred in the moderate warm habitats (T = 5.1-5.7) differentiated in terms of soil reaction (R), nitrogen availability (N), continentality degree (C), and moisture in particular.

Keywords: plant community, floristic diversity, biotopic conditions, bioindicator

## Introduction

The agricultural landscape of south-eastern Poland is dominated by arable land, while the grasslands are predominantly situated in the river valleys. They show high biodiversity arising from the interacting effects of various biogeographical components. Human activity, with the aims of land integration, soil reclamation, increased cattle stocking, intensive fertilization and automation of agricultural practices, has essentially been one of the major factors driving the change of biodiversity, i.e. towards landscape unification. Therefore, the valuation of nature in these areas is a vital element of the river valley environment assessment for the purposes of spatial planning and nature conservation.

In numerous regions in Poland it was found that the local floristic differentiation of the meadow communities of definite ecological spectra implies substantial variability of the field habitat conditions and anthropogenic factors. Recent years have been marked with a growing interest in usability of the phytoindicative procedures for meadow habitat valuation (Oświt, 1992; Barabasz, 1997).

The objective of the present research was to determine the floristic richness of permanent grasslands in the Huczwa river valley as well as evaluation according to Ellenberg *et al.*, (1992) phytoindicative method of some climatic conditions: insolation (L), temperature (T), continentality (C) as well as habitat – edaphic: moisture (F), acidity (R) and nitrogen content (N).

## Materials and methods

The phytosociological studies were carried out in the years 2001-2002 on the grasslands on hydrogenous soils in the Huczwa river valley (south-eastern Poland). The area under investigation is situated in the temperate climate zone with a continental influence. The total annual precipitation is approximately 600 mm. The studies covered the meadows and pastures

used by private farmers as well as those where cutting and grazing has ceased. There were made 79 relevés (phytosociological records) with the Braun-Blanquet method. The association diversity was evaluated on the basis of the total number of species distinguished and mean number of species in a relevé. The bioindicative assessment of the habitat conditions was performed for only those plant associations where at least the minimum number of 10 species was present in a relevé. According to the Ellenberg (1992) indicator value concept, the following mean values for each plot and for the different plant communities were calculated (on a 9-grade scale): L (relation to light), T (thermal relations), C (continentalism), R (soil reaction) and N (soil nitrogen availability), while for H (humidity) a 12-grade scale was used, including species frequency grades.

## Results and discussion

The grasslands in the Huczwa river valley are characterized by a rich floristic composition (Table 1). On the basis of the 79 phytosociological records, there were distinguished 9 associations of the *Phragmitetea* class (*Phragmitetum australis*, *Glycerietum maximae*, *Caricetum ripariae*, *C. acutiformis*, *C. rostratae*, *C. hudsonii*, *C. gracilis*, *C. vesicariae*, *Phalaridetum arundinaceae*), 2 associations and 1 community of the *Molinio-Arrhenathereta* class (*Scirpetum sylvatici*, the association with *Deschampsia caespitosa* and *Alopecuretum pratensis*) and association of the *Scheuchzerio-Caricetea nigrae* class (*Caricetum lasiocarpae*). However, species number in the discerned associations and in each relevé in a given association proved to be strongly differentiated. The highest species number was recorded in the associations *Caricetum acutiformis* (70) and *Caricetum gracilis* (66), whereas the lowest – *Phragmitetum australis* (16) and *Caricetum ripariae* (13). A mean species number in a phytosociological record ranged from 10.5 in the *Phragmitetum australis* up to 24.5 in *Caricetum lasiocarpae*. Similarly, in the studies of Kryszak *et al.*, (2005; 2006), the plant associations of the *Phragmitetea* class were shown as floristically poor, especially in the over-moist habitats. The following associations extended over the greatest area: *Caricetum acutiformis*, *C. gracilis* and *Alopecuretum pratensis* and the communities with *Deschampsia caespitosa*, whereas *Caricetum vesicariae* occupied the smallest area.

Table 1. Floristic diversity indices of the habitats distinguished.

Number of communities	Associations, community	Number of relevés	Total number of species	Mean in releve from-to mean	Number of communities	Associations, community	Number of relevés	Total number of species	Mean in releve from-to mean
I	<i>Phragmitetum australis</i>	4	16	9-12 10.5	VIII	<i>Caricetum vesicariae</i>	3	20	16-18 17,0
II	<i>Glycerietum maximae</i>	7	40	11-16 13.0	IX	<i>Phalaridetum arundinaceae</i>	5	22	13-15 14.0
III	<i>Caricetum ripariae</i>	3	13	10-14 13.0	X	<i>Scirpetum sylvatici</i>	3	22	18-20 19.0
IV	<i>Caricetum acutiformis</i>	17	70	7-24 17.5	XI	com. <i>Deschampsia caespitosa</i>	6	48	14-24 19.1
V	<i>Caricetum rostratae</i>	4	33	20-21 20.5	XII	<i>Alopecuretum pratensis</i>	6	41	17-27 20.2
VI	<i>Caricetum hudsonii</i>	5	28	14-18 15.6	XIII	<i>Caricetum lasiocarpae</i>	4	37	23-26 24.5
VII	<i>Caricetum gracilis</i>	12	66						

Explanations: I-IX – *Phragmitetea* class, X-XII – *Molinio-Arrhenathereta* class, XIII – *Scheuchzerio-Caricetea nigrae* class.

The phytoindicative assessment of habitats indicates that the associations and a community distinguished on the grasslands in the Huczwa river valley occurred in the habitats of similar values of thermal relation index (T) that ranged from 5.1 up to 5.7, i.e. a temperate warm habitat (Table 2). The *Phragmitetum australis* association characterized the habitats of the highest values of the following indices: humidity (H), soil reaction (R), nitrogen availability (N) and continentalism (C), while the community with *Deschampsia caespitosa* was recorded in the habitats of the lowest indices mentioned above. A range of mean values of the humidity index (H) for the plant associations distinguished appeared very broad (5.7-9.3), which implies strong differentiation of the humid conditions of the meadow habitats analysed.

Table 2. Range and mean values of light indicators (L), thermal relations (T), continentalism (C), humidity (H), soil reaction (R) and soil nitrogen availability (N) for various meadow plant communities.

Associations, community		L	T	C	H	R	N
<i>Phragmitetum australis</i>	range	6.7-7.0	5.0-5.2	4.3-5.0	8.6-10.0	7.0-7.1	7.1-7.6
	mean	3.6	5.1	4.7	9.3	7.0	7.4
<i>Caricetum acutiformis</i>	range	6.3-7.4	5.0-5.6	3.2-4.1	4.1-10.0	6.4-7.1	5.1-6.3
	mean	6.9	5.2	3.6	7.9	6.7	5.6
<i>Glycerietum maximae</i>	range	7.5-8.0	4.9-5.5	2.8-6.6	9.3-9.9	5.7-7.6	6.2-7.8
	mean	7.7	5.2	4.1	9.7	7.0	7.0
<i>Phalaridetum arundinaceae</i>	range	6.9-7.1	5.0-5.1	3.6-4.1	7.9-9.2	5.8-6.5	5.7-6.2
	mean	7.0	5.1	3.9	8.6	6.1	6.1
com. <i>Deschampsia caespitosa</i>	range	6.2-6.7	5.0-5.9	2.1-5.3	5.5-6.0	4.8-6.6	3.7-6.1
	mean	6.5	5.5	3.8	5.7	5.8	5.0
<i>Alopecuretum pratensis</i>	range	6.6-6.7	5.2-6.0	4.0-4.6	5.6-5.8	6.0-6.2	6.1-6.8
	mean	6.4	5.7	4.3	5.7	6.1	6.4

## Conclusions

On the grasslands in the Huczwa river valley, 12 plant associations and 1 plant community were distinguished. They constitute the recognized landscape and esthetic values, being at the same time the waterfowl refuge. The *Caricetum lasiocarpae* association belonged to the floristically richest, while *Phragmitetum australis* to the poorest ones.

The plant associations distinguished occurred in the temperate warm habitats (T from 5.1 up to 5.7), yet differentiated regarding the values of the following indices: soil reaction (R), nitrogen availability (N), continentalism (C) and humidity (H) in particular.

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# Floristic diversity and economic value of permanent grasslands in the Por river valley

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## Abstract

The agricultural value of fodder from floristically diversified permanent grasslands in the river Por valley (south-eastern Poland) was evaluated. Phytosociological assessment by means of Braun-Blanquet's method was performed on meadows and pastures. In total, 502 phytosociological records were made, among which 23 communities belonging to the following classes: *Phragmitetea*, *Scheuchzerio-Caricetea nigrae*, *Molinio-Arrhenatheretea* and *Plantaginetea maioris*, were distinguished. The grasslands yielded at a fairly low level and their economic value was differentiated (UVN from 0.1 up to 9.0).

Keywords: floristic diversity, plant communities, species, agricultural value

## Introduction

High biological richness is widely recognized as a major attribute of cultural landscapes that have botanically diverse grass and sedge associations, in addition to green landscape attributes (Kozłowski, 2002). Meadows and pastures account for approximately 22% of the cultivatable land in Poland, and in terms of their floristic diversity they constitute a natural fodder base which has not been fully utilized by conventional agriculture (Kozłowski, 1996; Młynarczyk *et al.*, 2001). The objective of the present work was to determine the agricultural value of forage produced on the floristically diversified permanent grasslands in the Por river valley.

## Materials and methods

The investigations were carried out on the permanent grasslands (private farmer's property) in the Por river valley (a left-bank tributary of the Wieprz river) situated in the south-eastern part of Poland. Prior to first-cut harvest (3rd decade of May and 1st June), a total of 502 phytosociological records were made with Braun-Blanquet's method and classified into an appropriate plant association according to Matuszkiewicz (2005). Phytosociological records were taken from 100-150 m<sup>2</sup> representative areas or, in exceptional cases, smaller (even below 10 m<sup>2</sup>) when an association occurred at very small area, e.g. in a drainage ditch. Agricultural value of most of the communities of meadows and pastures distinguished under this investigation was established on the basis of 127 plant samples collected randomly from a 2 x 1 m<sup>2</sup> area in the phytocenoses of each floristic unit. They comprised the material used for yield estimation and botanic-gravimetric analyses. The results provided by the botanic-gravimetric studies were also used to calculate the utility value number UVN according to Filipek (1973). The dry matter yield of the sward per 1 ha area was also calculated.

## Results and discussion

Currently, most meadows in the Por river valley are temporarily flooded – approx. 60% (*Caricetum lasiocarpae*, *Carici-Agrostietum caninae*, *Carex nigra* community, *Scirpetum silvatici*, *Cirsietum rivularis*, *Holcetum lanati*, *Calamagrostis epigejos* community, *Ranunculus acris* and *Lychnis flos-cuculi* community, *Alopecuretum pratensis*), permanently



wet – 20% (*Phragmitetum australis*, *Glycerietum maximae*, *C. acutiformis*, *C. gracilis*, *C. distichae*, *C. appropinquatae*, *C. rostratae*, *Phalaridetum arundinaceae*) or moist – about 20% (*Arrhenatheretum medioeuropaeum*, *Trisetetum flavescens*, *Poo-Festucetum rubrae*, *Lolio-Cynosuretum cristati*, *Rumici-Alopecuretum geniculati*, *Festuca arundinacea* community). The grasslands are situated on organic soils (about 60%) and on mineral soil (about 40%). In the Por river, the pronounced mosaic structure of phytocenoses was observed. 502 phytosociological records was made and analysed, 23 plant communities were distinguished and classified to the following: *Phragmitetea*, *Scheuchzerio-Caricetea nigrae*, *Molinio-Arrhenatheretea*, *Plantaginetea maioris*. The communities of the *Phragmitetea* and *Scheuchzerio-Caricetea nigrae* class occupied about 30% of all the meadow area, the class *Molinio-Arrhenatheretea* – 69.5% (communities of the order *Molinietales* – 30% and *Arrhenatheretalia* – 39.5%), with the others at 0.5%. The floristically richest proved to be the associations of the *Arrhenatheretalia* order: *Arrhenatheretum medioeuropaeum*, a typical variant – on average 33 species in one record; of the *Molinietales* order: *Holcetum lanati* – 29.1 species in one record; of the *Phragmitetea* class: *Caricetum distichae* – mean 30.0 species in one record; of the *Scheuchzerio-Caricetea nigrae* class: *Carici-Agrostietum caninae* – average 30.3 species in one record (Table 1). The plant communities discerned in the Por river valley included 274 vascular plant species. Among the vascular plants, there were determined 38 grass species, 17 legumes, 41 species of the *Cyperaceae* and *Juncaceae* family, 159 species of plants from the dicotyledenous plant class (a group of so called herbs and weeds), 14 trees and shrubs.

The floristic composition of meadows and pastures of the Por river valley appeared to be slightly poorer than that in the Dynowskie Plateau, where 363 vascular plant species were recorded, in that 42 grasses, 29 legumes, 41 sedges, rushes and field horsetails as well as 251 herbs (Wolański, 2006). In the sward of the meadows studied, there occurred protected, rare species in the Lubelszczyzna region and Poland, e.g. *Dianthus superbus*, *Dactylorhiza majalis*, *D. maculata*, *Epipactis palustris*, *Listera ovata*, some sedge species (*Carex lasiocarpa*, *C. davalliana*, *C. dioica*, *C. diandra*, *C. disticha*). The communities of *Phragmitetea* and *Scheuchzerio-Caricetea nigrae* classes provided forage of poor quality as indicated by UVN under 4, generally. Among the communities of the class *Phragmitetea*, the highest phytomass yields were obtained from rushes *Glycerietum maximae* (3.8-4.4 Mg ha<sup>-1</sup>). The poorest yield was recorded for short-sedge meadows of the *caricetum lasiocarpae* association – approximately 1 Mg ha<sup>-1</sup> hay in a cut, while the lowest UVN under 1 was shown by the hay of the *Caricetum rostratae* association. The latest researches indicate that silage produced from the meadow sward whose botanical composition is dominated by sedges (*Carex* spp.), is characterized by a higher nutritive value than hay (Żurek and Wróbel, 2006). As in this investigated valley, in the Dynowskie Plateau the highest sward dry matter content of the associations *Phragmitetea* class was obtained for the *Phragmitetum australis* and *Glycerietum maximae* communities. The utilization value numbers indicated that good quality sward was produced only by *Phalaridetum arundinaceae*, poor by *Glycerietum maximae*, while worthless by the other communities of the *Phragmitetea* class (Wolański, 2006). Of the associations of the *Molinietales* order, the meadow grass of the *Alopecuretum pratensis* was characterized by the highest yield (3.2-3.8 Mg hay in I cut) as well as the highest UVN (7.8-9.0). According to Miazga and Mosek (2001), foxtail meadows of the Kosarzówka valley also constituted a high productive potential. Among the phytocenoses of the *Molinietales* order in the Dynowskie Plateau, the highest yield was also noted for meadows of the *Alopecuretum pratensis* community (average 4.3 Mg ha<sup>-1</sup> DM) that scored high fodder value (Filipek, 1973). Meadow associations of the *Arrhenatheretalia* order were low-yielding but produced fodder of high utilization value, generally > 6.0. The highest yields were recorded for the meadows of *Trisetetum flavescens* community (1.8-3.1 Mg ha<sup>-1</sup>).

Table 1. Yields and forage value of meadow sward of each association.

Number of communities	Association community	Record number	Spps recorded	Mean species number	UVN-utility value number	Yield 1 cut in Mg ha <sup>-1</sup> (dry matter)
I	<i>P. australis</i> <sup>1</sup>	10	50	17.8	1.6-2.0 (1.8)	2.7-3.1 (2.9)
II	<i>G. maximae</i> <sup>1</sup>	20	90	14.2	3.8 (1.9)	3.8-4.6 (4.2)
III	<i>C. acutiformis</i> <sup>1</sup>	21	109	21.0	0.3-2.5 (1.5)	2.3-3.2 (2.7)
IV	<i>C. gracilis</i> <sup>1</sup>	43	148	21.0	0.4-3.4 (1.4)	2.0-3.6 (2.6)
V	<i>C. distichae</i> <sup>1</sup>	6	78	30.0	2.7-4.3 (3.5)	2.8-3.8 (3.3)
VI	<i>C. appropinquatae</i> <sup>1</sup>	16	114	22.7	0.3-2.1 (1.4)	1.5-2.0 (1.8)
VII	<i>C. rostrate</i> <sup>1</sup>	17	97	22.5	0.1-0.5 (0.3)	1.1-1.5 (1.3)
VIII	<i>P. arundinaceae</i> <sup>1</sup>	9	68	17.7	3.8-3.9 (3.9)	3.7-4.2 (4.0)
IX	<i>C. lasiocarpae</i> <sup>1</sup>	12	79	28.8	1.0-2.3 (1.8)	0.4-1.7 (1.1)
X	<i>C. caninae</i> <sup>1</sup>	10	87	30.3	3.9-4.7 (4.3)	1.1-1.9 (1.4)
XI	<i>C. nigra</i> <sup>1</sup>	19	122	24.2	1.9-5.1 (3.2)	1.0-1.5 (1.3)
XII	<i>S. sylvatici</i> <sup>1</sup>	8	75	24.0	2.0-2.7 (2.2)	1.8-2.0 (1.9)
XIII	<i>C. rivularis</i> <sup>1</sup>	29	125	27.9	0.2-4.3 (2.7)	1.2-3.2 (2.2)
XIV	<i>H. lanati</i> <sup>1,2</sup>	21	95	29.1	3.5-5.5 (4.3)	1.1-2.9 (2.2)
XV	<i>C. epigejos</i> <sup>1,2</sup>	10	73	20.2	3.4-4.2 (3.8)	1.4-2.2 (2.0)
XVI	<i>R. acris</i> <i>L. flos-culi</i> <sup>1</sup>	19	83	26.4	0.7-3.8 (2.5)	0.3-3.9 (1.5)
XVII	<i>A. pratensis</i> <sup>1,2</sup>	43	102	23.1	7.8-9.0 (8.1)	3.2-3.8 (3.5)
XVIII	<i>A. medioeuropaeum</i> <sup>2</sup>	85	125	33.0	5.1-8.1 (6.7)	1.8-2.8 (2.3)
XIX	<i>T. flavescentis</i> <sup>2</sup>	39	103	27.6	4.8-8.3 (6.5)	1.8-3.1 (2.3)
XX	<i>P. rubrae</i> <sup>1,2</sup>	40	133	24.3	6.0-7.7 (6.7)	1.4-2.7 (2.1)
XXI	<i>L. cristati</i> <sup>2</sup>	10	76	28.7	5.7-7.9 (6.7)	1.1-1.4 (1.2)
XXII	<i>R. geniculati</i> <sup>1</sup>	6	39	14.8	4.8-5.0 (4.9)	0.4-0.6 (0.5)
XXIII	<i>F. arundinacea</i> <sup>2</sup>	9	75	21.2	4.9-6.3 (5.4)	2.3-2.8 (2.6)

<sup>1</sup>organic soil, <sup>2</sup>mineral soil; \* range, \*\* mean; I-VIII *Phragmitetea* class, IX-XI *Scheuchzerio-Caricetea nigrae* class; XII-XXI *Molinio-Arrhenatheretea* class (XII-XVII *Molinietalia* order, XVIII-XXI *Arrhenatheretalia* order); XXII-XXIII *Plantaginetea maioris* class.

## Conclusions

Floristic composition on the investigated meadows and pastures reflects water conditions, soil trophic status and management. Generally, the yield was very low (0.3-4.6 Mg ha<sup>-1</sup>) due to the moisture conditions, lack of fertilization or inappropriate management. The UVN-value was less than 1 in some sedge communities, but up to 9 in some phytocenoses of the *Alopecuretum pratensis* association. Due to conservation values, ecological and aesthetic qualities the extensively used enclaves of all the plant communities in the Por valley should be maintained. However, the greater part of the meadows studied should produce good quality forage.

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# The effect of environmental factors on the occurrence of legumes in mountain grasslands

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## Abstract

Legumes play an important role on mountain grasslands especially under extensive management. They increase productivity of sward and improve fodder quality through improving the protein content and slowing down the lignifying process. The research was carried out in the Beskid Sądecki Mountains (Western Carpathians) at 400-1,000 m a.s.l. The occurrence and proportion of legume plant species was estimated on the basis of 312 phytosociological relevés made in grassland communities of *Molinio-Arrhenatheretea* and *Nardo-Callunetea*. From 16 legume species present in the survey, the most common (in 73% of relevés) was white clover (*Trifolium repens*) and red clover (*Trifolium pratense*) (in 62% of relevés). These species had also the highest average proportion. The main environmental factor influencing the occurrence of legumes was soil acidity. The effect of altitude and slope was also significant. Different species varied in their reaction to environmental factors. No effect of soluble phosphorus or potassium content in soil was found.

Keywords: mountain grasslands, legumes, habitat conditions

## Introduction

Legumes constitute an important element of grassland vegetation (Novoselova and Frame, 1992). Their presence in extensively managed mountain meadows and pastures is particularly important. In cases where no, or insufficient, fertilization is used they are the main source of soil enrichment in nitrogen compounds for improving sward productivity. They also positively affect the nutritive value of forage, particularly its protein concentrations and through considerably greater mineral content in these plants (Rochon *et al.*, 2004). Under extensive sward management, cutting is usually performed late and, the presence of a high proportion of legumes results in the lignification process being slowed down. The aim of this work was to make an assessment of legume occurrence on meadows and pastures in the areas with traditional agriculture and the dependence of this presence on selected environmental factors.

## Materials and methods

The research was conducted in the Beskid Sądecki Mts. (Western Carpathians), where extensive grassland management has been maintained to a considerable extent. Grasslands occupy slopes of various gradients and aspects, situated at different altitudes between 400 m and almost 1,000 m a.s.l. Depending on the location above the sea level mean temperature ranges between 7.7 °C and 3.6 °C and precipitation from 800 to 1100 mm. The soils are mainly leached and gleyed brown soils, and occasionally acid brown soils (mainly podzolized and typical podzols). In 2002-2005, 312 sociological relevés were made by the Braun-Blanquet method on selected 100 m<sup>2</sup> plots. The proportion of individual species in the sward was determined through conversion of Braun-Blanquet scale into area cover values. CANOCO Programme was used for statistical ordering of the relevés (PCA analysis).

## Results and discussion

Plant communities occurring on grasslands were classified into two phytosociological classes: *Molinio-Arrhenatheretea* and *Nardo-Callunetea*. Legumes were present mainly in meadow communities of the *Molinio-Arrhenatheretea* class. Occurrence of a total of 19 legume species was detected on the analysed grasslands (Table 1). They were present in 90% of the relevés. The share of this plant group in the area cover was relatively low, on average reaching 9%. The most frequently encountered species, which also had the greatest proportion of the botanical composition, were white clover (*Trifolium repens*) and red clover (*Trifolium pratense*). Tufted vetch (*Vicia cracca*) and meadow pea (*Lathyrus pratensis*) were also relatively frequent, but their proportion in the cover was smaller. Under conditions in central Poland these species are also among the most frequently encountered (Szozzkiewicz *et al.*, 1998). Other species appeared much more seldom, although some of them, e.g., zigzag clover (*Trifolium medium*) reached a considerable degree of area cover in individual patches.

Table 1. Frequency of occurrence and coverage (Braun-Blanquet scale) of legume species.

Species	Coverage			Species	Coverage		
	Frequency	Min	Max		Frequency	Min	Max
<i>Trifolium repens</i>	73	+	4	<i>Ononis arvensis</i>	4	+	3
<i>Trifolium pratense</i>	62	+	4	<i>Lathyrus sylvestris</i>	4	+	1
<i>Lotus corniculatus</i>	40	+	3	<i>Medicago falcata</i>	2	+	3
<i>Vicia cracca</i>	40	+	2	<i>Coronilla varia</i>	2	+	3
<i>Lathyrus pratensis</i>	17	+	3	<i>Medicago lupulina</i>	2	+	1
<i>Trifolium montanum</i>	13	+	3	<i>Trifolium pannonicum</i>	2	+	3
<i>Vicia sepium</i>	12	+	2	<i>Genista tinctoria</i>	1	+	1
<i>Trifolium dubium</i>	12	+	2	<i>Lotus uliginosus</i>	1	+	+
<i>Trifolium medium</i>	10		4	<i>Trifolium aureum</i>	1	+	1
<i>Anthyllis vulneraria</i>	8	+	3				

The main factors affecting legume proportion in the sward were soil reaction, the land slope and altitude above sea level (Figure 1). Occurrence of red and white clover was most strongly associated with increasing soil reaction value. It results from the relatively considerable soil and climatic requirements of these species, which are associated with slightly more intensively managed grasslands situated in the areas more favourable for cultivation (flat terrains situated on lower altitudes a.s.l, usually in the vicinity of farm buildings). Their sizeable proportion also results from these species seeding in the past. Due to their abundant occurrence, both these species play an important role in forage production on permanent grasslands in the investigated area, particularly that they are counted as among the most important forage plant group in Europe (Novoselova and Frame, 1992). At higher altitudes and on soils with lower pH, the legume proportion was considerably smaller and also other species apart from white and red clover were encountered here, particularly mountain clover (*Trifolium montanum*) and zigzag clover. Under these conditions, unfavourable for cultivation, even a small proportion of legumes may positively affect meadow productivity and their biodiversity. No dependence between the presence of these plants and soil-available

phosphorus was assessed. A lack of such a relationship was discovered also in other investigations conducted in Poland (Szozkiewicz *et al.*, 1998).

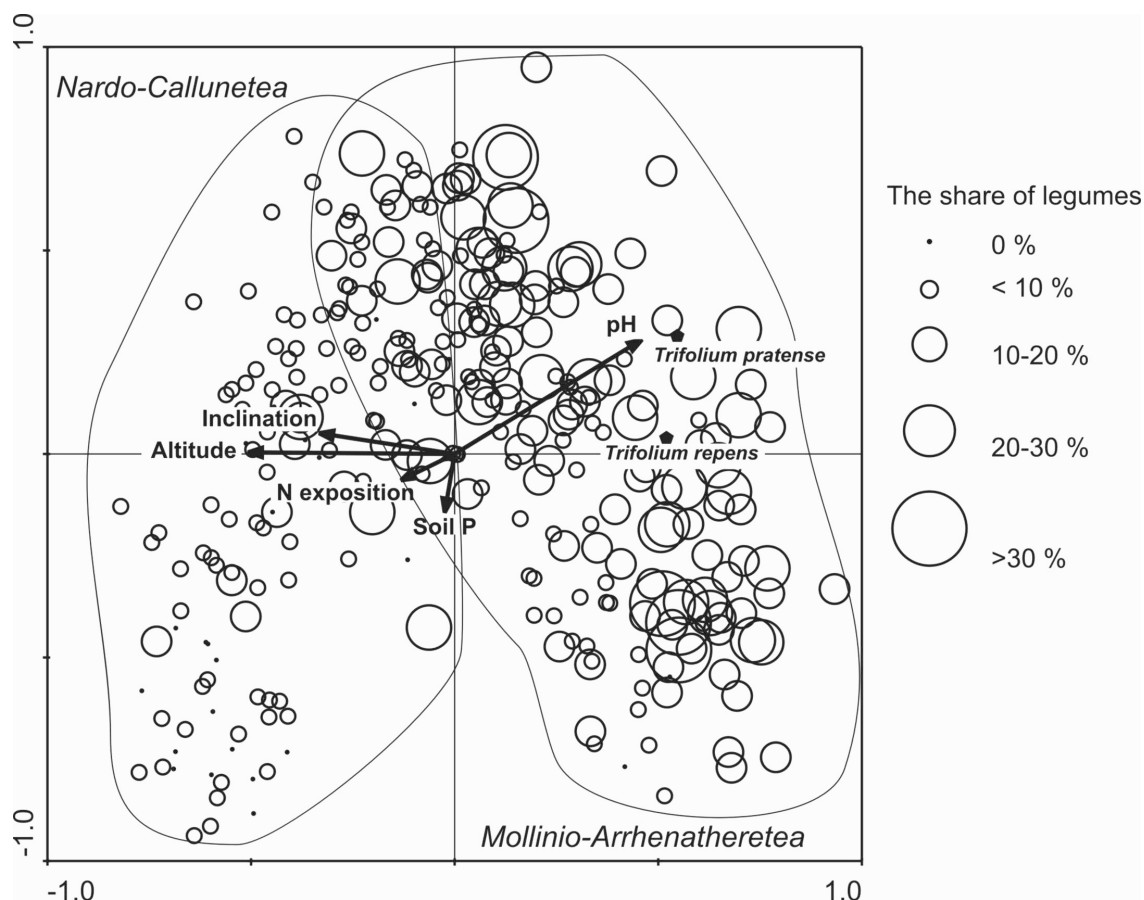


Figure 1. PCA ordination diagram of relevés, main legume species and environmental factors in relation to 1 and 2 axes.

## Conclusions

Occurrence of red and white clover, the main legume species, is connected with a habitat that has better agricultural usability and which is more intensively managed. The other species play an important role only in improving biodiversity. In the *Nardo-Callunetea* communities developed on marginal habitats, the share of legumes is minimal.

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## **Session 5B**

### **Low-intensity systems in future grassland production – benefits and risks**

# Performance of beef suckler cows and their progeny to slaughter on intensive and extensive grassland systems

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## Abstract

The performance of rotationally grazed, beef suckler cows and their progeny to slaughter on two lowland grassland Systems differing in stocking rate (SR) and fertilizer nitrogen (N) level was compared over eight years. The two Systems were 1) Intensive (INT): 0.58 or 0.72 ha cow<sup>-1</sup> unit, 211 kg fertilizer N ha<sup>-1</sup>, two silage harvests; and 2) Extensive (EXT) compatible with the EU, Rural Environmental Protection Scheme (REPS): 0.72 or 0.89 ha cow<sup>-1</sup> unit, 97 kg fertilizer N ha<sup>-1</sup> and one staggered silage harvest. Herbage dry matter digestibility both pre- and post-grazing was similar ( $P > 0.05$ ) for the two Systems, whereas herbage crude protein concentrations were generally lower for the EXT System. There was no difference ( $P > 0.05$ ) between the Systems in cow live-weight, body condition score or their changes, in calf live-weight gain pre- or post-weaning or in slaughter or carcass weight, kill-out proportion, estimated carcass gain, carcass conformation or fat score of the progeny. Efficiency of N use ha<sup>-1</sup> was higher on the EXT than INT System. In conclusion, similar individual animal performance can be expected in an extensive, lowland, grassland-based, suckler calf-to-beef System compatible with the EU, REPS as that attained in a more intensive System.

Keywords: beef suckler cow, cattle, extensification, grassland management systems

## Introduction

Extensive beef farming has been encouraged to reduce output and also to develop more sustainable production systems. Many farmers will now need to operate to lower input criteria than heretofore, as determined by environmental legislation and (or) stipulated in environmental schemes such as the EU Rural Environmental Protection Scheme (REPS). Consequently, grassland management systems will reduce in intensity and thus, there is a need to develop more extensive, REPS-compatible, systems of suckler beef production.

Within the context of a grassland-based, spring-calving, suckler beef production system, matching grass supply to animal requirements during the grazing season, while ensuring sufficient silage is produced for the indoor winter period, is largely achieved by balancing the use of fertilizer nitrogen (N) and stocking rate (SR) by altering the emphasis on harvesting grass by grazing or as silage. Whereas the use of two silage harvests has traditionally been advocated for moderately intensive suckler beef production systems, the considerable cost often associated with a second-harvest silage makes it necessary to consider replacing the two-harvest system with a simpler one-harvest system.

The objective was to compare two lowland, grassland management Systems, differing in SR and fertilizer N level, on the performance of rotationally grazed, beef suckler cows and their progeny to slaughter.

## Materials and methods

The study was carried out in Grange Beef Research Centre (Longitude 6°40'W; Latitude 53°30'N; Elevation 92 m a.s.l.) over a period of 8 years (1997-2004) on a permanent grassland site, sub-divided into paddocks. Swards consisted predominantly of *Lolium perenne*. The soil type was a moderately well drained Brown Earth with a clay loam texture. Long-term mean

(s.d.) annual rainfall and daily air temperature were 849 (81.6) mm and 9.1 (0.64) °C, respectively.

Spring-calving (27 March (s.d. 26.7)) cows comprising 0.5 to 1.0 late-maturing continental breeds mated to continental breed sires were used. Heifer progeny were slaughtered at 607 (s.d. 26.8) days of age except in Years 3 and 4 (retained for breeding). Male progeny were slaughtered as steers at 717 (s.d. 20.4) days in Years 1 and 2 and subsequently as bulls at 453 (s.d. 25.3) days of age. The two Systems were 1) Intensive System (INT): SR of 0.58 (bull) or 0.72 (steer) ha cow<sup>-1</sup> unit (cow plus progeny to slaughter), 211 kg (s.d. 12.1) fertilizer N ha<sup>-1</sup>; two silage harvests; and 2) Extensive System (EXT): SR of 0.72 (bull) or 0.89 (steer) ha cow<sup>-1</sup> unit, 97 (s.d. 5.5) kg fertilizer N ha<sup>-1</sup> and one silage harvest.

A total of 57 and 148 kg ha<sup>-1</sup> fertilizer N (generally urea 46%N) was applied to the grazing area of EXT and INT, respectively. On the silage harvesting area, the mean N application rate was 110 and 80 kg ha<sup>-1</sup> for first and second harvests, respectively, with further applications of 55 and up to 20 kg ha<sup>-1</sup> following the final harvest of herbage for INT and EXT, respectively. Phosphorus and potassium fertilizer application rates were based on soil test analyses. Slurry produced during the winter indoor period was returned to the area harvested for silage at a rate proportional to the Systems SR. In INT, the first precision-chop silage harvest (for progeny) was taken on 24 May (s.d. 3.4) and the second on 4 August (s.d. 7.2). In EXT, half of the area was cut at the same time as INT (24 May) with the remainder deferred and conserved 17 days later (10 June, s.d. 2.8).

Grazing commenced for the cows and calves on 21 April (s.d. 16.1). Calves were weaned and accommodated indoors at 218 (s.d. 28.4) days of age. During their first winter period, the heifers and steers were offered grass silage *ad libitum* and 1.0 kg of concentrate (barley-based) daily, then turned out to pasture (6 April (s.d. 7.0)) for a second grazing season and subsequently housed on 3 (s.d. 28.8) and 27 October (s.d. 12.0), respectively. Bulls remained indoors post-weaning. For the finishing period, grass silage was supplemented (commenced at pasture for heifers) with approximately 3, 5 and 5 kg concentrate per head daily over a final 54 (s.d. 30.9), 129 (s.d. 10.5) and 238 (s.d. 17.1) days for the heifers, steers and bulls, respectively. Cows were offered grass silage (deferred and second harvest) *ad libitum* plus a mineral-vitamin supplement daily in winter, except at first calving when, additionally, 1.5 kg of concentrate was offered *post-partum* until grazing commenced.

Pre- and post-grazing sward height and/or herbage mass was measured using a rising-plate meter and by cutting  $\geq$  four strips per paddock, respectively, in the first three years, and herbage nutritive value was determined in Years 1 and 3. Animal live-weight, body condition score (BCS), carcass weight, carcass grades and feed analysis were determined as described by Drennan and McGee (2004). Nitrogen balance ha<sup>-1</sup> was estimated assuming an N content of live-weight of 28 g kg<sup>-1</sup>. Data were analysed using general linear models with terms in the model for System, year, dam genotype, parity, sire and gender, where appropriate. Birth day was included as a covariate.

## Results and discussion

On the cow grazing area the Systems did not differ ( $P > 0.05$ ) in sward height or mass pre- (~11.7 cm; ~2300 kg) or post-grazing with the exception that post-grazing values were lower (5.7 vs. 6.3 cm; 424 vs. 555 kg,  $P < 0.05$ ) for INT than EXT in Year 1. Herbage *in-vitro* dry matter digestibility (DMD) both pre- (~756 g kg<sup>-1</sup>) and post- (~653 g kg<sup>-1</sup>) grazing was similar ( $P > 0.05$ ) for both Systems. Herbage crude protein (CP) concentrations were lower ( $P < 0.01$ ) post-grazing in Year 1 (152 vs. 180 g kg<sup>-1</sup>) and lower ( $P < 0.01$ ) both pre- (159 vs. 205 g kg<sup>-1</sup>) and post- (141 vs. 172 g kg<sup>-1</sup>) grazing in Year 3 for EXT than INT, respectively. On the area grazed by the yearlings, Systems did not differ ( $P > 0.05$ ) in sward pre- and post-grazing heights, mass, DMD or CP concentration. The mean DMD of the first, deferred and



second harvest silages were 716 (s.d. 39.2), 689 (s.d. 37.1) and 674 (s.d. 41.6) g kg<sup>-1</sup>, respectively. Systems did not differ ( $P > 0.05$ ) in cow live-weight, BCS or their changes or in progeny pre- and post-weaning live-weight gain, slaughter or carcass weight, kill-out proportion, estimated carcass gain, carcass conformation or fat score (Table 1). Estimated N surpluses were 209 and 95, and 206 and 93 kg ha<sup>-1</sup> for INT and EXT steer and bull production systems, respectively, with corresponding N-use efficiencies of 0.10 and 0.17, and 0.12 and 0.20.

Table 1. Animal performance on the Intensive and Extensive grassland management Systems.

Grassland Management System	Intensive	Extensive	s.e.m.
<b>COW</b>			
Live-weight and changes (kg)			
Housing	588	587	4.6
Winter change	-44	-42	2.8
Grazing season change	+82	+82	2.4
Body condition score and changes (units 0-5)			
Housing	2.7	2.7	0.05
Winter change	-0.31	-0.32	0.048
Grazing season change	+0.22	+0.16	0.045
PROGENY daily live-weight gain (g)			
Pre-weaning	1027	1009	13.4
Post-weaning			
Indoor winter 1			
Heifers	386	391	14.8
Steers	441	401	73.3
Bulls	1208	1238	35.4
Grazing season 2			
Heifers	894	885	15.3
Steers	808	796	60.6
Indoor winter 2			
Heifers	886	838	38.1
Steers	864	901	95.2
Carcass gain per day of age (g)			
Heifers	465	465	4.1
Steers	514	517	17.8
Bulls	734	739	11.4

## Conclusions

Similar individual animal performance and higher efficiency of N use ha<sup>-1</sup> can be expected in an extensive grassland-based suckler calf-to-beef System compatible with the EU, REPS as that attained in a more intensive System comprising of both a higher SR and fertilizer N application.

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# The effect of outdoor cattle keeping in winter season on water and soil quality

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## Abstract

In the foothills of the Jeseníky Mountains (Czech Republic) the influence of cattle kept outdoors in winter (one sheltered area; the other one non-sheltered) on soil and surface water quality was monitored during 2004-2007. Soil samples were taken at depths of 2-30 cm and 30-60 cm at the cattle wintering place and on the adjacent pastures, in spring and autumn. Water samples were taken from a stream above and below the wintering place every other month. The cattle wintering had a significant influence on the increase of phosphorus (P) content in the soil at depth of 2-30 cm and on soil potassium (K) content at both depths. The content of soil K was significantly higher at the sheltered winter locality. The content of calcium was significantly lower in the 30-60 cm soil layer as a result of wintering. The stream waters were not contaminated by nitrite and nitrate nitrogen (N). The content of ammonium N was higher in the first two years of monitoring (more in the sheltered stand), but in the last year it did not exceed the pollution limit. The content of P showed a tendency to increase in the sheltered wintering area in the last year.

## Introduction

Cattle breeding and grazing is one of the main farming activities in the less favourable areas. The year-long keeping of cattle on pastures is rather common in America and Australia, and is of interest from an economic point of view. But year-long keeping of animals builds up a risk of high concentrations of phosphorus (P), potassium (K) and ammonium (NH<sub>4</sub>) compounds and losses to the environment. Thus, it is important to protect the soil, and surface and underground waters from pollution from cattle-wintering areas, especially in accordance with the Nitrate Directive (Council of Europe Directive No. 676/91). The aim of this study is to assess the impacts of winter and year-long cattle pasturing on soil and surface water quality.

## Materials and methods

The effect of outdoor cattle outdoor on the quality of soil and surface water has been studied at the Research Institute for Cattle Breeding in Rapotin, Czech Republic since 2004. The monitored areas were situated in the foothills of Jeseníky Mountains, in areas at 340-390 m above the sea level. The average annual temperature was 6.2 °C and the average annual rainfall was 700 mm. The two localities chosen were Salas (S) – for sheltered wintering, and Annov (A) – non-sheltered wintering area. There were 60-80 cows and group of calves in the herd, with a stocking rate of 0.9-1.1 LU ha<sup>-1</sup> at both localities. The cattle wintering place (shelter) and the feeding place were located at 30-50 m from the stream. In Annov the wintering place was moved every year. Samples of water were taken from the upper (U) and lower (L) streams, the upper sampling was situated in forest. The average water flow in the Salas stream was 0.11 m<sup>3</sup> s<sup>-1</sup>, and the Annov stream reached a water flow rate of 0.06 m<sup>3</sup> s<sup>-1</sup>. The indicators of quality monitored were: N-NO<sub>2</sub><sup>-</sup>, N-NO<sub>3</sub><sup>-</sup>, N-NH<sub>4</sub><sup>+</sup>, P as well as the value of biochemical oxygen demand and dichromate value; the sampling was taken in two-month intervals (the water was sampled 20-times in total). Both localities are situated on sandy-loam soil, sub-unit luvi-cambisols. The soil was sampled directly at the cattle wintering place (the

highest animal concentration) and at the adjacent pastures (check sampling) in spring (April) and in autumn (October). The samples were taken from the depth of 2-30 cm and 30-60 cm (soil was sampled 7 times). The soil samples were analysed for the following: content of inorganic nitrogen ( $N_{in}$ ), content of available nutrients (P, K, Ca) by Mehlich III method. The results were evaluated using SPSS for Windows v. 13.0.

## Results and discussion

The various forms of nitrogen affect water quality differently. Nitrate is the most prevalent soluble form affecting freshwater and has generally been considered the most 'troublesome' contributor to acidification of waters and eutrophication, both of which result in major changes to aquatic biodiversity and community structure (Nash and Haygarth, 2005). The streams were not contaminated by  $N-NO_2^-$  (limit of pollution  $0.05 \text{ mg l}^{-1}$ ) or by  $N-NO_3^-$  (limit of pollution  $7 \text{ mg l}^{-1}$ ) according to the pollution indicators of surface water in the monitoring period of time. Ammonium-N is dangerous and toxic for fishes; its limit for surface water is  $0.5 \text{ mg l}^{-1}$ . The contents of  $N-NH_4^+$  were also higher than the acceptable limit in the upper streams in the 2004-2005 period. This was caused by the high content of  $N-NH_4^+$  in rainfall (according to the data provided by the Institute laboratory) and 'throughfalls' – rainfall in the lower parts of forest vegetation which contain higher concentration of pollution than the open areas. Cattle grazing (wintering) also had the impact on higher pollution in this period. The highest concentrations were measured in August (S U  $0.89$ , S L  $1.23$ , A U  $0.91$ , A L  $1.02 \text{ mg l}^{-1}$ ) and October 2004 (S U  $0.69$ , S L  $0.91$ , A U  $0.75$ , A L  $0.85 \text{ mg l}^{-1}$ ). Since December 2005  $N-NH_4^+$  contamination has decreased under the pollution limit. Elevated concentrations of phosphorus in streams, rivers and lakes may contribute to eutrophication (Pierzynski *et al.*, 2000). The limit of pollution concentration ( $0.15 \text{ mg l}^{-1}$ ) has been exceeded five times in the Salas locality under the wintering place (pasture) and twice in the Annov locality (at both sampling places) in that period of time. Figure 1 shows that P pollution in the Salas locality has increased in 2007, which could be caused by the high phosphorus concentration in the soil (Rzonca *et al.*, 2006) that has been splashed in the water. From this point of view non-sheltered wintering that is moved every year is better for surface water quality.

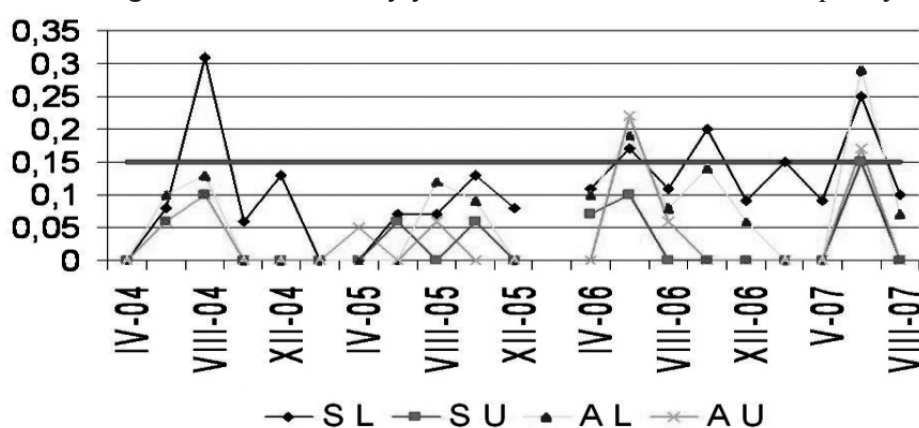


Figure 1. The course of phosphorus concentrations ( $\text{mg l}^{-1}$ ) in lower (L) and upper (U) monitored streams for Salas (S) and Annov (A) from April 2004 to October 2007.

The value of biochemical oxygen demand and dichromate value serves for estimation of organic pollution. The pollution limit by biochemical oxygen demand ( $6 \text{ mg l}^{-1}$ ) was exceeded only twice in the Salas locality and once in Annov locality. The pollution limit by dichromate value ( $35 \text{ mg l}^{-1}$ ) was exceeded three times in both localities under the wintering area as a result of cattle that was kept there. The values of nutrient content in soils are in Table 1.

Table 1. The basic agrochemical properties of soil (mean of years 2004-2007).

Locality	Monitored area	Depth (m)	N <sub>in</sub> (mg kg <sup>-1</sup> )	P (mg kg <sup>-1</sup> )	K (mg kg <sup>-1</sup> )	Ca (mg kg <sup>-1</sup> )
Salas	pasture	0.02-0.30	7.80	37	218	2163
		0.30-0.60	5.20	18	126	1733
	wintering place	0.02-0.30	24.48	164	1191	1543
		0.30-0.60	15.16	132	395	959
Annov	pasture	0.02-0.30	14.24	50	181	2177
		0.30-0.60	9.97	29	141	1822
	wintering place	0.02-0.30	26.54	57	533	1752
		0.30-0.60	14.03	29	262	1095
ANOVA	Locality	0.02-30	0.26	2.49	6.63*	0.09
(F – ratio)	Monitored area		2.14	4.34*	22.46**	3.87
	Season		3.70	1.85	0.23	0.10
	Locality	0.30-0.60	0.00	0.96	1.25	0.45
	Monitored area		3.20	1.29	8.29**	13.45**
	Season		6.16*	1.79	1.64	0.27

\*  $P < 0.05$  \*\*  $P < 0.01$ .

The cattle had no influence on the increase of nitrogen content in the 2-30 cm layer, but showed a tendency to increase in the deeper layer. The content of inorganic N was significantly higher in autumn than in spring in the deeper layer (there was tendency to this increase in the 2-30 cm layer). There were no significant differences of P content in different localities and seasons. The cattle had a significant influence on the increase of P content in the 2-30 cm layer. The content of K was significantly higher in Salas locality than in Annov and there was a significantly higher K content in both monitored depths caused by cattle wintering. The content of calcium showed a tendency to decrease at the depth of 2-30 cm and it was significantly lower in 30-60 cm depth as a result of cattle wintering.

## Conclusions

Wintering place could contaminate surface water by phosphorus and ammonia nitrogen. On the basis of the results achieved we can conclude that changing the wintering place every year and following the proper farming routines (high bedding, regeneration of grass turf in the spring) can eliminate the risk of the above-mentioned contamination. Wintering was shown to cause an impact in terms of an increase of potassium and phosphorus content and a decrease of calcium content in the soil.

## Acknowledgements

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# Simulating the impact of dry climatic years on grassland utilization and production from beef cattle farming systems based on permanent pasture

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## Abstract

The simulation of grassland production and utilization at farm scale allows evaluating the feed self-sufficiency of the farming system for different management strategies and for different climatic years. We investigated the impact of the frequency of years unfavourable to grassland production, such as 2003 that was characterized by drought and high temperatures. We used the whole-farm simulation model 'Sebien', which reproduces the steady-state functioning of grassland-based beef suckler systems. The model is based on the interfacing of an animal sub-model with a vegetation sub-model and a management sub-model, and was calibrated for suckler farms studied in the upland areas of the Auvergne region of France. Simulation results indicated that without changing the management rules, in 2003 the amount of herbage harvested was reduced in proportion by 0.54 and grass quality was markedly reduced compared with a 'normal' year without climatic extreme events, such as 2000, leading to a dramatic decrease of farm feed self-sufficiency. The response of the model to the increasing frequency of year 2003 was linear. Changes in harvesting rules allowed to contain the loss of herbage harvested to 0.34 in proportion and to maintain grass quality. This study highlights the interest of building management rules that take into account extreme climatic years.

Keywords: forage system, permanent pasture, beef cattle, drought, feed self-sufficiency, simulation

## Introduction

Climate change will probably produce increased frequency of heat stress and droughts. These might negatively affect crop yields and livestock beyond the impacts of mean climatic variables (Easterling *et al.*, 2007). In particular, beef suckler systems that rely exclusively on permanent pastures, as can be found in upland areas of Europe (e.g. Massif Central, France), might be very sensitive to extreme climatic events such as the drought and heat stress observed in spring and summer 2003.

Models are useful tools for understanding and quantifying interactions between the components of the farming system and for testing alternative scenarios. A whole-farm simulation model (Sebien) reproducing the steady-state functioning of grassland-based suckler farms was developed (Jouven and Baumont, 2008). In this study we used 'Sebien' to analyse the impact of extreme climatic events that might reduce grass production and to investigate management scenarios that could help coping with a growing frequency of such climatic events.

## Materials and methods

'Sebien' predicts the functioning of the farm at a daily time scale, with simulation runs lasting 1 to 15 years. Paddocks, animal groups and animal categories within groups are the management units. The model is made of three sub-models which interact at multiple time scales. The grassland resource sub-model (Jouven *et al.*, 2006) predicts grass growth and quality at the paddock level, from soil quality, vegetation functional traits and climatic data. The animal



sub-model (Jouven *et al.*, 2007) calculates selective intake at pasture from the biomass and digestibility of plant parts. It also predicts weight gain and milk production from energy intake, for each animal type within each group. The management sub-model (Jouven and Baumont, 2008) comprises a strategic component (management plan) and a tactical component (management rules). Herd management works mainly on a pre-planned schedule. Utilization of paddocks is also planned, but can be revised at fixed dates depending on the amount of herbage available to graze. Management rules adjust feed availability for the herd, through grazing rotations, hay harvests, and supplementation with forage and concentrate to achieve production objectives (calf weight at sale, cow body condition score at calving).

Based on a real farm typical of the upland regions of the Massif Central, we simulated a virtual farm composed of 30 ha of permanent grasslands of medium productivity, and of a herd of 26 cows and 10 replacement heifers. This corresponds to a medium stocking rate of 1.1 LSU per ha. Almost half of the surface (14 ha) was planned to be harvested as hay at heading stage in spring. Calving occurred on January 15 and the calves are weaned and sold at 9 months at a weight of 325 kg. The simulations were run using the weather data recorded at the INRA research station of Marcenat (France, altitude 1100 m), where average annual precipitation reaches 1200 mm per year and mean daily temperature is around 7 °C. The model was first run on the weather data recorded between 1995 and 2002 to adjust management rules in order to maximize feed self-sufficiency.

To study the impact of drought and heat stress we chose the year 2003 that was characterized by a 0.53 reduction in rainfall between April and June and by a 3 °C increase in average temperature between May and June compared to the average values of last forty years. First, we analysed the performance of the farming system in 2003 and the impact of increasing the frequency of this climatic event from once every six years up to once every two years, without any change in management rules. For that purpose we built artificial 12-year climatic series combining the year 2003 and the year 2000, that was characterized by the absence of extreme climatic events. Then, we tested if adaptations of management rules, with a particular focus on those dealing with hay harvests, could improve farming system performance when unfavourable years as 2003 are frequent in the climatic series.

To evaluate the performance of the farming system with a particular focus on feed self-sufficiency, we considered the global outputs of the model. These were the forage harvested or grazed, and the hay and concentrate purchased, expressed per livestock unit (LSU).

## Results and discussion

Feed self-sufficiency of the simulated farm was almost fully achieved on the reference climatic series as very little hay was bought, and only 45 kg of concentrate per LSU were used, mainly for the calves (Table 1). The variability between years was more pronounced for the amount of forage harvested than for the amount of forage grazed, since management rules secure the utilization of the paddocks for grazing.

The simulated impact of the climatic conditions in 2003 was disastrous on the herbage harvested (-0.54 in proportion) and negative on the herbage grazed (-0.20). The important decrease in harvested herbage was explained by a lower biomass production at the first cut in spring (1.8 vs. 3.5 t DM ha<sup>-1</sup>) and by the absence of a second cut in summer, when all the paddocks were needed for grazing. As a consequence feed self-sufficiency decreased dramatically (Table 1). The concentrate purchased increased in response to the decrease in herbage quality (0.56 vs. 0.63 for the organic matter digestibility of the harvested herbage).

The results for the 'normal' climatic year 2000 are in the range of those achieved with the reference series. The response of the model to the increasing frequency of year 2003 in the artificial climatic series was a linear decrease of the herbage production and of farm feed self-sufficiency. Indeed, the results for the climatic series combining the years 2003 and 2000 in



equal frequency were close to the average of the results obtained for the two years separately. This indicates that in the simulation there was little or no residual effect of the year 2003 on the following years, which could be explained by the favourable weather conditions for grassland production in autumn 2003.

Climatic conditions in 2003 led to reduced growth in relation to drought and to earlier maturation of herbage, and increased losses of biomass through senescence in relation to heat stress. To cope with the reduction in herbage quality and availability in 2003-years, we tested the following adaptations in management rules: i) early first-cut at the beginning of heading (instead of at heading); ii) harvest of barn-dried hay or big-bale silage, which reduces to 3 the numbers of dry days needed for harvesting (4 days needed for hay); iii) threshold of 1.5 t DM ha<sup>-1</sup> for first-cut and of 1 t DM ha<sup>-1</sup> for second-cut harvests (instead of 1.7 t DM ha<sup>-1</sup> for both cuts). Applying these three rules together increased the production of harvested herbage by 0.45 and its quality (0.64 vs. 0.56 for the organic matter digestibility) in 2003-years. Feed self-sufficiency was markedly improved for 2003-years, but not for 2000-years (Table 1). In a context of climate change, this highlights the interest of taking into account extreme climatic years rather than average ones to set management rules.

Table 1. Evaluation of the performance of the farming system through the herbage produced and the feed purchased (in t DM LSU<sup>-1</sup>) for different series of climatic years.

	Reference years 1995-2002	With management rules of reference years			With adapted rules for harvesting		
		Dry years (2003)	'Normal' years (2000)	½ Dry ½ Normal	Dry years (2003)	'Normal' years (2000)	½ Dry ½ Normal
Produced herbage (t DM.LSU <sup>-1</sup> )							
Harvested	1.71 ± 0.31	0.78	1.54	1.16	1.13	1.38	1.25
Grazed	2.59 ± 0.10	2.06	2.63	2.34	2.15	2.63	2.39
Purchased feeds (t DM.LSU <sup>-1</sup> )							
Hay	0.08 ± 0.12	1.73	0.23	0.98	1.11	0.27	0.69
Concentrate	0.045 ± 0.009	0.177	0.041	0.109	0.074	0.033	0.053

## Conclusions

This study highlights the interest of a simulation approach to investigate the ability of farming systems to cope with climate changes. The use of the model allows testing different scenarios involving farm structure and management of the farming system. The results of simulations should provide a basis for further analysis with farmers and extension services.

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# Impact of mixed grazing on animal performance and moorland vegetation

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## Abstract

As part of a project on economic and environmentally sustainable moorland management practices, two systems-scale studies, one site in Wales and one in northern England, are being undertaken on the impact of mixed grazing on animal performance and moorland vegetation. Long-term sheep grazing studies were previously conducted at both sites to investigate the impact of Environmentally Sensitive Area stocking rates prescribed for restoration or maintenance of heathland. Cattle grazing was introduced at each of the sheep stocking rates on commencement of the new studies. Changes in liveweight and body condition score for cattle at the England site were acceptable for the class of cattle used; however, the performance of the lambs at the restoration stocking rate has improved, whereas lamb performance on the maintenance stocking rate has declined. At the Wales site, the performance of the cattle has varied between years; however, the performance of the sheep has been unaffected. Changes in the flora and fauna have also occurred at both sites.

Keywords: animal performance, cattle, lamb, heather, semi-natural rough grazings

## Introduction

Upland heaths in the UK are of international conservation significance. These heaths are traditionally managed by extensive grazing systems that maintain the semi-natural plant communities and provide valuable income to farms in the hills and uplands. Increasing numbers of sheep in the second half of the 20th century resulted in overgrazing and a subsequent degradation of upland heaths, with dwarf shrubs and bog-mosses being replaced by competitive grasses. To counteract these trends, agri-environment schemes were introduced in the UK in the late 1980s encouraging farmers to reduce sheep numbers. More recently, schemes have attempted to reintroduce cattle grazing to hill and upland sites. Cattle and sheep graze differently and have different selection strategies when grazing heath communities. However, farmers will only adopt grazing management prescriptions that are financially viable, practical and do not compromise animal production or welfare.

As part of a large, multi-disciplinary project to determine both economic and environmentally sustainable moorland management practices, two systems-scale studies were undertaken to determine the impact of mixed grazing on animal performance and moorland vegetation.

## Materials and methods

The four-year studies started in 2002 and were undertaken at ADAS Pwllpeiran (Mid Wales) and ADAS Redesdale (Northumbria, northern England). Prior to the studies commencing, both sites were running long-term investigations with sheep, to examine the impact of Environmentally Sensitive Area (ESA) grazing prescriptions on the restoration or maintenance of semi-natural hill vegetation.

At ADAS Pwllpeiran, the site comprised 72 ha of *Nardus stricta*-dominant grassland, formerly dwarf shrub heath degraded by past heavy grazing. The main objective was to reduce the abundance of *N. stricta*, to restore dwarf shrubs such as *Vaccinium myrtillus* and *Calluna vulgaris*, and maintain the diversity of other plant species. The treatments were low sheep (1.0 sheep ha<sup>-1</sup>), high sheep (1.5 sheep ha<sup>-1</sup>), low sheep plus cattle (1.0 sheep +

0.5 cattle ha<sup>-1</sup> for two months in summer) and cattle-only grazing (0.5 cattle ha<sup>-1</sup> for 2 months). Treatments were replicated over three blocks. Welsh Mountain ewes (40 kg liveweight) and yearling Welsh Black heifers (approx 460 kg liveweight) were used. Cattle were turned out for grazing for two months each summer.

At ADAS Redesdale, the study was carried out on 103 ha of degraded wet heath. The vegetation comprised a mosaic of *C. vulgaris*, *Molinia caerulea*, and *Juncus* spp. plant communities. The area had been split into two paddocks and, from 1995-2002, grazed by Scottish Blackface sheep (55 kg liveweight) at 1.5 and 0.66 ewes ha<sup>-1</sup> (minus 25% October-February). The management objective was to reduce the abundance of *M. caerulea* to facilitate the restoration of dwarf shrubs and the mosaic of wet heath plant communities. From 2003-2006, the paddocks were split again and sheep grazing regimes continued, but non-lactating, autumn-calving Simmental x Holstein and Belgian Blue x Holstein mature cows grazed from mid-June each year. Grazing treatments were low sheep (LS; 0.66 ewes ha<sup>-1</sup>), low sheep plus cows (LSC; 0.66 ewes plus 0.75 cows ha<sup>-1</sup>), high sheep (HS; 1.5 ewes ha<sup>-1</sup>) and high sheep plus cows (HSC; 1.5 ewes plus 0.75 cows ha<sup>-1</sup>). Cows were removed at the first signs of their grazing *C. vulgaris* in mid-summer, and vegetation condition checked by measuring the length of five laminae of *M. caerulea* at 30 random locations per paddock.

At both sites, liveweight and condition score of cattle were recorded at the start and end of the grazing period. Birthweights, and weaning weights were recorded for all lambs. All livestock data were analysed using REML (Genstat). For vegetation assessments, presence and top cover of all plant species, litter and bare ground were recorded pre-treatment and at the end of the experiment, and at intervals during the experimental period. Records were made of presence and top cover of all plant species, litter and bare ground, local shoot frequency and grazing index of key species at each site.

## Results and discussion

At ADAS Pwllpeiran, animal performance varied more between years than between treatments. There was no significant effect of treatment on daily liveweight change (DLC) of cattle ( $P > 0.05$ ), although there were consistent trends. In the first year (2003), which was the first time that cattle had been introduced to these areas for many years, there was high biomass of vegetation, which had been neglected by sheep, and cattle performance was very similar ( $P > 0.05$ ) on both treatments (0.49 vs. 0.54 kg day<sup>-1</sup> for cattle-only and mixed grazing, respectively). In subsequent years, cattle performance was consistently *ca.* 25% lower under mixed grazing. There was an effect of year (SED = 0.07,  $P < 0.001$ ) with gains in 2005 being higher than in other years. This is consistent with the vegetation results. With more palatable species being present in the sward in September 2004 and a very warm summer with adequate rainfall, coupled with lower sward heights, it would be expected that more palatable, leafy and nutritious vegetation would have been available, increasing nutrient intakes and leading to improved performance. Overall, gains in cattle liveweight were sufficient for growing heifers, calving at 3 years old. There was no effect of treatment on lamb weaning weights ( $P > 0.05$ ), although there was a significant year effect (SED = 0.73,  $P < 0.001$ ). Lamb performance appeared to be improving year on year (24.1, 25.5, 27.4 kg respectively), which again would be expected from the small but significant vegetation changes recorded. There was a significant interaction between year and treatment on ewe weaning weights (SED = 1.40,  $P < 0.05$ ). In 2006, ewes on the low stocking rate had a significantly greater liveweight at weaning than ewes on the high stocking rate (41.0 kg vs. 38.1 kg respectively), with ewes on the mixed grazing regime having intermediate values. In previous years, there was a similar trend ( $P > 0.05$ ) between ewes on the two sheep-only treatments.

The overall grazing effect on the plant community was on both frequency and cover, indicating some change in community structure as well as differences in the amount of shoot

biomass. However, the effects were mostly small in magnitude, reflecting the low dynamism of this vegetation. In 2004, cover of productive species (*Festuca. ovina*, *Agrostis. capillaris*, *M. caerulea*) was greater, and that of less-productive species (*Juncus squarrosus*, *Carex binervis*) was lower. This suggests a more productive year, supported by trends in vegetation height. For *M. caerulea*, at least, this was also related to reduced grazing intensity. *Vaccinium myrtillus* showed no overall change, despite being widely grazed.

At ADAS Redesdale, with the exception of 2005, cattle from the HSC treatment had lower daily liveweight change than cattle on the LSC treatment (SED = 0.21,  $P < 0.05$ ). The higher sheep stocking decreased cattle DLC by an average of 210 g. There was considerable variation between years and, in 2006, cattle on LSC treatment had DLC of 1.19 kg day<sup>-1</sup> compared with HSC of 0.64 kg day<sup>-1</sup>. Gains in cattle liveweight were sufficient for non-lactating cows. However, grazing periods were shorter after the first two years, being 9, 10, 6 and 4 weeks in 2003-2006, respectively. This was attributed to the biomass available at the start of each season and throws doubt on the sustainability of an annual mixed grazing regime at these stocking densities. Lambs were weaned at an average of 127 days. Weaning weights were significantly higher for lambs from the LS treatments whether cattle were present or not (SED = 0.67,  $P < 0.001$ ). Growth rates from birth to weaning were 236, 222, 208 and 211 g day<sup>-1</sup> for treatments LS, LSC, HS and HSC respectively ( $P < 0.001$ ). Daily liveweight gain from birth to marking at an average 25 days of age (361, 336, 316 and 299 g day<sup>-1</sup> respectively;  $P < 0.001$ ) suggested that much of this difference was due to increased milk yields of ewes. The inclusion of cattle had little effect on lamb performance with the exception of 2006 when there was an apparent reduction in performance at the lower stocking rate. Sheep stocking rate had a significant effect on ewe tupping weight in all years (SED = 0.68,  $P < 0.001$ ) with an average reduction of about 6 kg per ewe from the higher stocking rate. The effect of cattle grazing with ewes was small overall but there were indications that ewe performance was improving with time when grazing with cattle.

The most important effect was the increase in grazing of *M. caerulea*, with reduction in its cover and exposure of the moss and litter layer after cows were introduced. However, neither *C. vulgaris* nor any other species had spread apart from *Juncus* spp. in HSC. Therefore, there was no evidence of restoration after four years of mixed grazing, although suitable conditions appeared to have been created. This contrasts with the sheep-only paddocks where *M. caerulea* increased (especially in HS) at the expense of *C. vulgaris* cover. However, *C. vulgaris* frequency was increasing under the low grazing levels in LS, which might be beneficial in the longer term.

## Conclusions

Historically, sheep grazing at high stocking densities have resulted in a reduction of dwarf shrubs and replacement by other species such as *M. caerulea* and *N. stricta*. At low stocking densities, sheep are much less damaging to both established *C. vulgaris* and new seedlings and can also reduce *N. stricta* by facilitating competition from broad-leaved grasses. Summer grazing by cattle is potentially beneficial for moorland regeneration by reducing competitive grasses. Cattle are particularly effective for reducing *M. caerulea*, but their effectiveness for managing *N. stricta* appears to be more variable and dependent on plant community composition. Performance of both traditional and continental breeds can be satisfactory although welfare issues can arise with continental breeds under inclement weather conditions.

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# Evaluating an organic low input grassland dairy system farming on permanent pastures in eastern France (Vosges lowland)

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## Abstract

Since late 2004, a spring-calving dairy cow herd has been managed to farm 80 hectares of permanent pastures with an extended grazing period (March to November) and winter feeding based on hay and very few concentrates. Using a systems experiment, the main objective consists in designing and evaluating an environmentally friendly system. Results from the first two years of running have been analysed. Despite a rather good working of the forage system, reproductive performance of cows was dramatically impaired. Some ways of improvement were planned regarding the position of the calving season in the year. Spring grazing management, and especially fodder supply for the cows during this period, were also reconsidered. Some compromises between grassland production, biodiversity and some other environmental issues still remain to be designed.

Keywords: dairy herd, permanent pasture, environmental objectives, biodiversity, prototyping

## Introduction

Social demand for sustainable agriculture and studies such as the FAO's *Livestock's long shadow* (Steinfeld *et al.*, 2006) should lead livestock systems to be more respectful of environmental issues. In such a context, research needs to search for (re-)designing and evaluating agricultural systems. In North-Eastern France, a research team is prototyping two complementary organic livestock farming systems based on (i) respect of land capabilities and resources preservation, (ii) low use of inputs, and (iii) limitation of exchanges between systems.

So as to achieve their environmental sustainability, several goals have to be assigned to these systems, such as resource preservation (water, air and energy) and increased use of environmental components for agricultural production (biodiversity and soil fertility).

Experimenting at farming system level combined with modelling might suit well that prototyping purpose. In this paper, we present some first results of the assessment of an organic low input Grassland dairy System, so called GS. The GS is one of the two prototyped systems in the Mirecourt experimental unit.

## Materials and methods

The systems were designed by using a method adapted from Vereijken (1997), which requires the defining of a set of objectives and decision rules for the operating system. The decision rules aim at making the system management more explicit. Evaluating the achievement of the different objectives is based on a farm-scale experiment including two different aspects: (i) the farming system operation level, with animal and grassland production issues, and (ii) the environmental impact of the farm on different components (water, air, soil, energy, pasture floristic composition). But it is also important to evaluate decision rules, which have to take into account several diversified objectives.

The GS aims at producing milk while respecting and enforcing its environmental properties, mainly by maximizing the grazing of permanent pastures. We have assumed that grazing is

the cheapest method of livestock feeding. In September 2004, the GS relied on a spring-calving 37-cow dairy herd managed to farm permanent pastures (78.5 ha). It was decided that cows (from two breeds, Holstein and Montbéliarde) would have the main part of their lactation between March and November, during the grazing season. This decision aimed at producing lower cost milk. Besides, winter feeding was decided to be based on hay only. But, in such a configuration, a good management of the reproductive performance of the dairy cows became crucial to get a lasting herd.

At farm operation level, we assess the production results which stem from the running of the system, guided by the decision rules. For that purpose, criteria involving both animal and grassland production aspects were used. The grass cover was monitored weekly using grassmeter height measurements. The grazing management was analysed with a special attention for the spring period in order to highlight the relationship between grass budget, milk yield and reproductive performance of the cows. A pasture-use calendar gathering the field management practices data allowed the calculation of the stocking rate, the grass budget, and also the estimation of the animal grass intake. Moreover, the harvested hay was accurately accounted for in order to refine the balance between grass production, animal feed supply and forage stores sufficiency.

Concerning the environmental issues we only investigated two components at the very beginning of the system experiment: plant grassland diversity and soil fertility. Changes in pasture composition since the organic farming conversion in 2004, nutrient cycling and water and air qualities still remain in progress.

## Results

The spring grazing management relied on a turnout date as early as possible according to the available grass (late March and early April respectively in 2005 and 2006). Consequently, the topped pastures were 10 ha large in 2005 but only 6 in 2006. The day and night rotational use of 21 ha in 2005, and 17 ha in 2006, led to a mean spring stocking rate of 2.5 cows and 2.8 cows ha<sup>-1</sup> respectively, until the grazing was enlarged by extended on to previously cut hay fields. Despite the grass growth being slightly lower in the spring of 2005 than in 2006 (34 vs. 39 kg DM ha<sup>-1</sup> day<sup>-1</sup> respectively), the mean regrowth duration on pastures before grazing stayed the same, about 35 days from May to late June. In return, the grazing days in hand were very different: much higher in 2005 than in 2006 (22 vs. 12 days from April to mid-June) giving more reliability to the animal supply in 2005. However, the daily milk yield was the same in both 2005 and 2006: 23 to 24 kg cow<sup>-1</sup> from turnout till the summer grazing enlargement. But dramatically impaired reproductive performance was observed during these first two years (Fiorelli *et al.*, 2007), especially for the Holstein cows and for the year 2006: in 2005, 65% of the cows submitted for breeding were pregnant at the end of the open period (Holstein: 52%; Montbéliarde: 81%) but they were only 27% in 2006 (Holstein: 17%; Montbéliarde: 33%). A thorough analysis of individual reproductive performance suggested that the latest calving cows (April to May) encountered more difficulties in successful insemination, probably because of the energy shortage at the beginning of their lactation stimulated by the higher grass intake.

Assuming this impaired reproductive performance, an increase of the culled cows number led to a marked decrease of the mean lactating cows number between 2005 and 2006 (36 to 31 cows), especially during the grazing period. Nevertheless, the cumulative harvested area and the total hay yield stayed exactly the same at the GS level through the two years (189 and 192 Mg DM). Owing to no grazing supplementation was required during the summer periods, GS faced with oversized hay stores: whilst about 3.0 Mg DM LU<sup>-1</sup> were harvested at the farm scale, only 2.0 to 2.3 Mg DM were needed to supply animal intake from October to June.



## Discussion

It seems very difficult to design another grazing management, for instance through increased regrowth duration before grazing, without considerable changes in the pasture composition. Therefore, in a first step, we preferred to test another method of improvement: making the calving period earlier from mid-January, while keeping a feeding supplementation of the cows with hay until June. Most of the cows will then begin their lactation with a forage based diet (hay and aftermath without any concentrates) resulting, hopefully, in only a limited energy shortage. In addition, the supply of hay during the whole the spring period should limit the grass intake effect. A new balance needs to be defined between grazing and cutting strategies to ensure the best reproductive performance.

On the other hand, in order to ensure the herd longevity, first through the stock numbers, 13 non-pregnant but lactating cows were kept during winter 2006-2007. Such a decision rule allowed the keeping of a 35-cow dairy herd, to limit the hay stores excess, and to test new lifetime profiles that may be relevant for the livestock system suitability.

Not only does this experimental design enable us to raise questions concerning the general operation of a given system, but it is also a relevant basis to develop experiments which aim to answer disciplinary questions associated with a larger application.

The determination of species composition of permanent pasture vegetation from the set up of the GS has shown differences in terms of plant species richness and diversity according to previous farming practices. Intensive grazing without cutting decreased plant species richness and diversity compared to several cuts a year and a low grazing intensity (Gaujour *et al.*, 2007). Thus, farming practice background must be taken into consideration to determine the impacts of such a GS on plant composition. A vegetation experiment was also set up at farm scale to distinguish GS effects, previous farming practices effects and effects of farm landscape characteristics. The landscape is implied in dispersal of seeds through habitat fragmentation and connectivity and contains important seeds sources such as field boundaries.

## Conclusions

The operating of this prototype reveals the difficulty of the design of a livestock farming system because of the various processes to combine and to manage: ensuring herd longevity, maintaining pasture production and enhancing biodiversity, while controlling the nutrient cycling to avoid losses to waterways or the atmosphere. But the systems experiment run at the Mirecourt INRA unit is an approach which allows both systemic and analytical investigations and so gives opportunities to different research teams to collaborate on a multidisciplinary project, primarily designed to come with the agriculture changes towards a better environmental sustainability.

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# Economy of beef production on extensive pasture in a nature conservation area

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## Abstract

In 2004-2006, an experiment was carried out in which the effectiveness of beef production from heifers on extensive pasture situated in a nature conservation area near Brody Experimental Station of Agricultural University Poznań (Poland) was analysed. The established pasture was intended for 24-hour grazing of heifers. Experimental animals (112-118 heifers) were purchased in spring, grazed during the vegetation season lasting 182-204 days and then sold in November. In the grazing herd two groups of heifers were presented: younger (7-9 months) and older (17-19 months). Each year the daily gains of heifers were measured by weighing and the evaluation of the economy of beef production was performed. Additionally, the botanical composition of the sward and dry matter (DM) yield of pasture were estimated. During the pasturing season daily gains of animals ranged from 449 to 736 g, depending on the heifer age group and the pasture yield in experimental years. Extensive grazing of heifers appears to be an economically justified method, as indicated by the values of return of beef production inputs, for agricultural utilization of nature conservation areas.

Keywords: economic efficiency, extensive pasture, heifers, nature conservation area

## Introduction

In low-input grassland management, grazing with beef cattle is considered an appropriate method of sward utilization (Isselstein *et al.*, 2005). In an attempt to find balanced, environmentally and landscape-friendly forms of management of permanent grassland, an experiment was carried out on the possibilities for beef production from the pasture in a nature conservation area. The objective of this work was to evaluate the economy of beef production of heifers grazing this pasture in two age groups.

## Materials and methods

During 2004-2006, studies were carried out to evaluate the effectiveness of beef production from heifers on extensive pasture in a nature conservation area near the Brody Experimental Station of Agricultural University Poznań (52°26'N, 16°18'E). The pasture was intended for 24-hour grazing using heifers. The area of pasture (42.4 ha) was divided into 6 paddocks. Experimental animals (112-118 heifers) of the Polish Holstein-Friesian breed were purchased in spring from among animals which were selected as unsuitable for milk production and grazed during a grass-growing season lasting 182-204 days. In the grazing herd two groups of heifers were presented: Y – younger (7-9 months) and O – older (17-19 months). One person was employed to service the herd for 4 hours per day. Forage from the pasture was the only feed the heifers received. Fresh water was brought to the animals twice a day and they also received straw and mineral supplements. At the end of the pasturing season in November all the animals from the herd were sold. The pasture had been established in 1988 on a muck soil formed from low-bog peats ( $\text{pH}_{\text{KCl}}$  6.5,  $\text{N}_t$  – 0.68%,  $\text{P}_2\text{O}_5$  – 53.2 mg 100 g<sup>-1</sup>,  $\text{K}_2\text{O}$  – 37.0 mg 100 g<sup>-1</sup>,  $\text{Mg}$  – 7.8 mg 100 g<sup>-1</sup>). Mineral fertilization of pasture was restricted to 50 kg ha<sup>-1</sup> N, 50 kg ha<sup>-1</sup>  $\text{P}_2\text{O}_5$  and 90 kg ha<sup>-1</sup>  $\text{K}_2\text{O}$  applied in spring. Every year, the botanical composition of the sward using the point method and dry matter yield of pasture were estimated. The daily

liveweight gains of heifers were measured by weighing. The evaluation of the economic effectiveness of beef production using the method of differential computation and calculation of total costs (Ströbel, 1987) was performed. All calculations were made on the basis of 2007 prices, assuming purchase and sale prices at the same level of 4.3 PLZ kg<sup>-1</sup> (1 PLZ = 0.27 €).

## Results and discussion

Changes in botanical composition of the sward were observed over successive years of pasture utilization (Figure 1). The dominant sward component was *Poa pratensis*. Its proportion ranged from 32% to 44%. *Trifolium repens* occupied 8-11% depending on the year. *Agropyron repens* and *Taraxacum officinale* occurred in significant quantities: 11-15% and 8-10%, respectively. An increase in the number of species in the pasture sward was observed and during the years of the investigation this ranged from 36.5 (on average) in 2004 to 44.5 in 2006. The grazing groups had no significant effect on the botanical composition of the pasture sward.

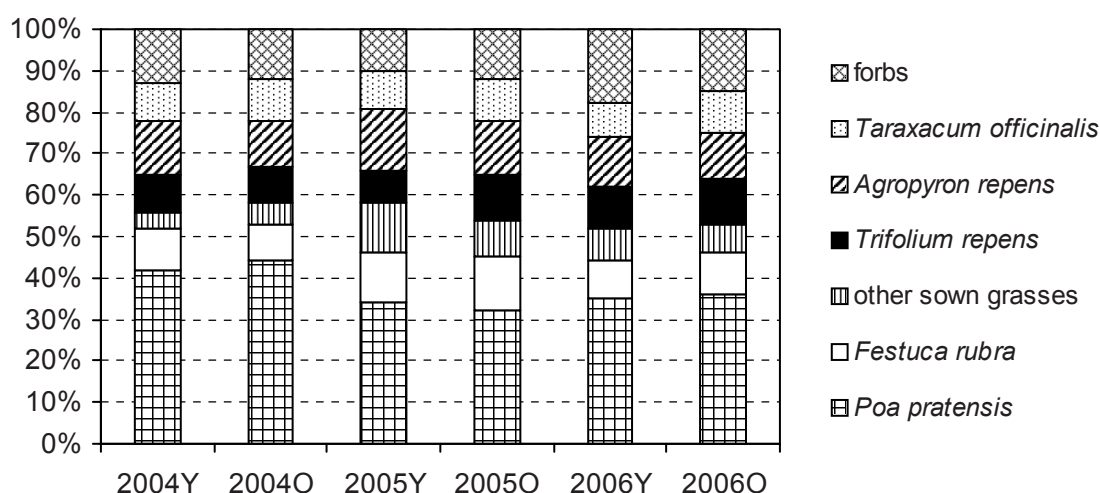


Figure 1. Changes in botanical composition of the sward over successive years of utilization.

The dry matter yield of pasture showed small variations between individual years and grazing groups, and ranged from 4,860 to 5,450 kg ha<sup>-1</sup>. The highest liveweight gains (736 g day<sup>-1</sup>) were recorded in 2005 in the older group of heifers, and the lowest (449 g day<sup>-1</sup>) in 2004 in the younger group of heifers (Table 1). Pavlu *et al.* (2001) found that heifers can maintain a good individual performance through selective grazing when the herbage on offer exceeds the demand. In each year of the study the higher gains occurred in the older group of grazing animals.

Table 1. Characteristics of heifers beef production on extensive pasture.

Item	Unit	2004		2005		2006	
		Y	O	Y	O	Y	O
Group of heifers							
Number of heifers in the herd	heifers	64	54	50	62	54	58
Buying weight of heifer – mean±SE	kg	275±34	397±23	245±52	419±42	263±25	377±19
Selling weight of heifers – mean±SE	kg	360±26	520±30	359±49	553±49	389±24	519±21
Grazing period	days	189	197	204	182	196	196
Daily liveweight gain – mean±SE	g	449±45	624±58	559±61	736±73	642±32	722±58
Labour engagement per grazing period	lh	418.7	344.1	364.3	403.0	378.0	406.0
Average capital engaged in production	PLZ	59,290	75,276	44,691	84,066	49,819	76,262

The production of beef ranged from 213.1 to 357.0 kg ha<sup>-1</sup> and was higher in the older group of heifers (Table 2). The efficiency of beef production on a low-input pasture is most affected by the purchase price. Following Poland's accession into the European Union in 2004 the economy of beef production on extensive grasslands was increased. Additionally, since 2004 the farmers have had the opportunity to receive financial compensation when they integrate nature conservation objectives into their grassland systems. Wytrzens and Neuwirth (2004) reported that there is a correlation between field-based subsidies and the use of grassland, its multifunctionality and nature conservation. Productivity factors expressed in economic terms confirmed the importance of the better use of pasture sward by older animals. The highest return of beef production inputs were recorded in 2006 in both groups of heifers (Table 3).

Table 2. Natural and financial productivity of production inputs on extensive pasture.

Production input	Unit	2004		2005		2006	
		Y	O	Y	O	Y	O
Pasture	beef kg ha <sup>-1</sup>	213.1	308.3	287.3	336.0	317.1	357.0
Labour	beef kg lh <sup>-1</sup>	12.7	18.9	15.4	20.2	17.7	19.9
Capital	beef kg 100 PLZ <sup>-1</sup>	7.4	7.6	10.3	8.4	11.1	9.2
Pasture	PLZ ha <sup>-1</sup>	916.1	1,325.7	1,235.5	1,445.0	1,365.9	1,534.9
Labour	PLZ lh <sup>-1</sup>	54.6	81.2	66.3	86.7	76.2	85.6
Capital	PLZ 100 PLZ <sup>-1</sup>	31.8	32.5	44.1	36.3	47.5	39.6

Table 3. Return of beef production inputs on extensive pasture.

Production input	Unit	2004		2005		2006	
		Y	O	Y	O	Y	O
Pasture	PLZ ha <sup>-1</sup>	424.7	765.2	738.1	888.3	868.5	977.0
Labour	PLZ lh <sup>-1</sup>	31.3	52.9	45.6	59.2	54.4	60.5
Capital	PLZ 100 PLZ <sup>-1</sup>	20.7	24.8	32.4	28.3	36.2	31.2

## Conclusions

Extensive grazing of heifers appears to be an economically justified method, as indicated by the values of return of beef production inputs, of agricultural utilization of nature conservation areas with positive effects on biodiversity of the grassland. During the pasturing season daily liveweight gains of animals ranged from 449 to 736 g depending on the heifers age group and year of investigations. Higher economical effects were reached by grazing of the older group of heifers.

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# The nutritional value of pasture forage for sheep in the Krkonoše Mountains National Park

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## Abstract

The nutritional value of pastures in the marginal mountain regions of the Krkonoše Mountains National Park (Czech Republic) situated at an average altitude of 1,235 m (Zadní Rennerovky), was determined. This protected landscape area provides important ecological and environmental functions. Extensive grazing by sheep helps to preserve rare, protected and endangered species of plants (including endemics), and also helps restoration of pasture biodiversity. The objective of this experiment was to determine the production of plant biomass and *in-vivo* digestibility of the above pastures with sheep. Over the years 2001-2004 the average dry matter forage yield was 4,988 kg ha<sup>-1</sup>. Digestibility of organic matter was 62.7% and 52.6% for young and mature forage, respectively. The results of the present experiment showed that the mountain grasslands have a potential as grazing areas for sheep. Grazing can also act as a tool to preserve the biodiversity of this type of mountain grassland.

Keywords: sheep grazing, production of biomass, nutritional value, digestibility

## Introduction

Pastures, including unique plant communities, were established in the 16-17th centuries in the Krkonoše Mountains National Park as a product of human activities. Species composition of pastures evolved along with continuous management. Until the Second World War pastures were mown in mid-summer and grazed by cattle for the rest of the vegetation season. However, subsequent abandonment of pastures has resulted in decreased species richness (Krahulec *et al.*, 2001). Sheep grazing represents an alternative to traditional management of pastures in the Krkonoše Mountains. The advantage of sheep grazing is that, due to their lower body weight, sheep harm the grassland less than cattle. The park performs important ecological and environmental functions, and has a high aesthetic value and sport and recreational potential. Extensive sheep grazing helps to preserve rare, protected and endangered species of plants (including endemics), and restoration of pasture biodiversity. Re-establishing and conservation of grassland biodiversity is of vital importance in some West European countries in which forage has been produced very intensively over recent decades (Kramberger and Gselman, 2000).

The objective of this experiment was to estimate the production of plant biomass in these pastures, and to determine its *in-vivo* digestibility with sheep.

## Material and methods

The experimental pasture (area 3 ha) is situated in the territory of the Krkonoše Mountains National Park (Czech Republic), in the locality of Zadní Rennerovky (50°41'54"-50°41'57"N; 15°39'58"-15°40'10"E) at an altitude of 1,220-1,250 m. The pasture is located on a southern slope with acidic coarse-grain gneiss and mica-schist bedrock. Mean temperatures during July and January are of 11 °C and -6 °C, respectively, with an average yearly precipitation of 1,100 mm (Krahulec *et al.*, 2001). Abandonment of the meadows since the 1960s has caused domination by *Polygonum bistorta*, which are classified as independent phytocenological units (associations of *Polygono-Deschampsietum flexuosae* and *Junco filiformis-Polygonetum*



(Pecháčková and Krahulec, 1998). From 1990 to 2000 the area was grazed by sheep. From 2001 is an observed area under a mulching regime (once per year at the end of July).

Samples of biomass were taken from twelve small enclosures (0.5 x 0.5 m), situated in the pasture, twice during a vegetation period, i.e. in the middle (July) and in the end (the beginning of October) of the grazing period, during 2001 to 2004. During the first grazing period two averaged forage samples were collected; i.e., forage A (date of harvest 9th of June) and forage B (date of harvest 16th of August). Forage samples were evaluated for the digestibility of organic matter, crude protein and crude fibre with four sheep, using fattened merino rams weighing approximately 85 kg. Chemical compositions were analysed according to the AOAC (1990).

Data were statistically analysed using the General Linear Model (GLM) procedure of Statistical Analysis System (SAS Institute, 2003). Statistical differences were determined by the Scheffe test.

## Results and discussion

The average forage dry matter (DM) yield for four years (2001-2004) was 4,988 kg ha<sup>-1</sup>. Forage DM yield (above-ground biomass) was 5,230, 5,190, 4,380 and 5,150 kg DM ha<sup>-1</sup> in 2001, 2002, 2003 and 2004, respectively. The nutrient concentrations (g kg<sup>-1</sup>) of both forages (A and B) are presented in Table 1. Higher contents of DM and crude fibre were found in sample B. Differences in chemical composition correspond to the date of sampling. The grazing season is short in this altitude, with growth starting immediately after the snow melt (mid-May), and lasts until October. Vegetation reaches the phenological and production maximum in July (Krahulec *et al.*, 2001).

The digestibility coefficients (%) of nutrients in forages A and B are given in Table 2. Maturity of forage had an important effect on the digestibility of nutrients. Digestibility of organic matter and crude protein was significantly lower in sample B ('mature' forage) than A ('young' forage). Fibre content in forage is a suitable indicator of its nutritional value, and is negatively correlated with digestibility of organic matter and digestibility of crude protein (Mrkvička and Veselá, 2002). In comparison with Fiala and Gaisler (2000) the present study shows lower DM yield of forage. They found DM yield of forage of 6,660 kg ha<sup>-1</sup> for grassland (from 1997 to 1999; mulching once per year at 15th of July) in the foothills of the Jizerské Mountains.

Table 1. Nutrient content (g kg<sup>-1</sup>) of forage (A and B) in dry matter.

	Dry matter (g kg <sup>-1</sup> )	Crude protein (g kg <sup>-1</sup> )	Ether extract (g kg <sup>-1</sup> )	Nitrogen-free extract (g kg <sup>-1</sup> )	Crude fibre (g kg <sup>-1</sup> )	Ash (g kg <sup>-1</sup> )
Forage A	180	39	5	79	44	13
Forage B	425	56	10	185	147	27

Table 2. Digestibility coefficients (%) of nutrients of forages (A and B).

	Organic matter (%)	Crude protein (%)	Crude fibre (%)
Forage A	62.7	68.0	56.6
Forage B	52.6	55.4	50.2
<i>P</i> <	0.05	0.01	ns

## Conclusions

Over the years 2001-2004 the average dry matter forage yield was 4,988 kg ha<sup>-1</sup>. Digestibility of organic matter was 62.7% and 52.6% for 'young' and 'mature' forage, respectively. The



results of the present experiment showed that the mountain grasslands have a potential as grazing areas for sheep. Grazing can also act as a tool to preserve the biodiversity of this type of mountain grassland.

### Acknowledgements

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# Effects of natural pasture on body condition in fit Standardbred geldings

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## Abstract

The aim of this study was to evaluate the effect of keeping fit Standardbred geldings ( $n = 6$ ) on natural pasture on body weight (BW) and body condition scores. From July 7 to August 6 2007 the geldings were kept on a range consisting of open grass areas (natural pasture) and forest. The geldings were weighed before and after pasture and body condition was estimated. There was a significant increase in the BW ( $497 \pm 58$  vs  $539 \pm 55$  kg) and a significant decrease in the sum of body scores ( $6.3 \pm 1.2$  vs  $5.2 \pm 1.6$ ) following pasture. There was a poor correlation ( $R^2 = 0.30$ ) between the change in the sum of scores and the change in BW before and after pasture. In conclusion, this study indicates that natural late-summer pasture may not be adequate to maintain body condition in fit Standardbred horses and that BW might be a poor indicator of body condition.

Keywords: horses, pasture, body weight, body condition, crude protein

## Introduction

In Sweden, pasture (cultivated as well as natural) is used in horse management only to a limited extent. This is due to the cold climate and the lack of grass growth during some months, but there is also a lack of knowledge of how pasture can affect body condition and health. Nevertheless, pasture is used as 'vacation' or rehabilitation when athletic horses are supposed to recover from injury. The aim of this study was to evaluate the effect of keeping fit, healthy Standardbred geldings on natural pasture for one month on body weight (BW) and body condition score and to evaluate if changes in these variables are correlated.

## Materials and methods

Six healthy, Standardbred geldings (5-8 years) in training were used. All horses had been racing and were near race condition. For 2-3 months prior to the pasture period the horses were fed individually. The diet consisted of 10 kg grass haylage (timothy and meadow fescue, 69% dry matter (DM)), 2 kg oats, 1.5 kg molassed sugarbeet pulp, 0.75 kg soy bean meal, 0.25 kg brewers yeast and 1 kg lucerne pellets per 490 kg BW ( $110 \text{ MJ ME day}^{-1}$ , Crude Protein (CP)  $140 \text{ g kg}^{-1} \text{ DM}$ ). Water was offered from automatic pressure valve water bowls (flow  $4.6\text{--}8 \text{ l min}^{-1}$ ). All horses were dewormed with Noromectin® and had passed a dental examination/treatment during the prior 2-3 months.

From 7 July to 6 August 2007 the geldings, plus another three geldings (in similar condition) were kept on a 17 ha range, located 50 km north of Stockholm, Sweden (Lat  $59^{\circ}72'N$ , Long  $17^{\circ}81'E$ ), consisting of open grass areas (natural pasture) and forest. The open grass area corresponded to approximately 40% of the total range and was dominated by timothy (*Phleum pratense* L.), smooth-stalked meadow-grass (*Poa pratensis* L.), cocksfoot (*Dactylis glomerata* L.), meadow fescue (*Festuca pratensis* L.), dandelion (*Taraxacum web* L.), Yarrow (*Achillea millefolium* L.), cow parsley (*Anthriscus sylvestris* L.) and junipers (*Juniperus* L.). The forest consisted mainly of spruce (*Picea abies*) and aspen (*Populus tremula*). At the start of the pasture period parts of the grass areas were over-mature. A salt block and two 250 l troughs of water were available.

All horses were weighed (U137, Vågsspecialisten, Sweden) before and after pasture and body condition was assessed. The body condition score system for back, hindquarters and belly is shown in Figure 1. Half scores were used if the observation did not fit to a full score. Scores for ribs were: 1 p = not visible, 2 p = visible during motion, and 3 p = easy to observe at rest. The intention was that scores for back, hindquarters and ribs (BHR) could indicate changes in amounts of body tissue, and for belly line changes in volume of the viscera/intestinal fill (positive correlations). Fresh faecal grab samples were collected before pasture (1-3 samples per horse and day for 2-3 days) and on the last day on pasture (from 1-2 defecations per horse) for analysis of CP content. Values are presented as means  $\pm$  standard deviation of the mean. Data were subjected to a paired t-test. The *P* value for significance was  $< 0.05$ .

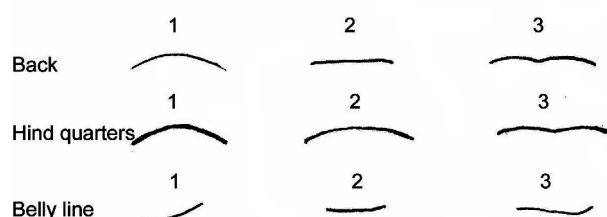


Figure 1. Score evaluation for back, hindquarters and belly line. Back and hindquarter conformation was evaluated from behind the horse (distance 7 and 20 cm on both sides of the spine, respectively) and belly line from the side of the horse (line from front legs to hind legs).

## Results

At the end of the period the pasture was either very short cut (less than 2 cm), short cut (2-10 cm) or very long (30-70 cm). The majority of the pasture was not lush and the plants were partly withered and yellow. The precipitation during this period was 39 mm, which is approximately 55% of the normal mean (30 years, SMHI, 2007) and might have reduced the growth of the pasture. The mean ambient temperature was 16.5°C, which is equal to the normal mean.

There was a significant increase in the mean BW ( $497 \pm 58$  vs.  $539 \pm 55$  kg) and a significant decrease in the mean sum of body scores for BHR ( $6.3 \pm 1.2$  vs.  $5.2 \pm 1.6$ ) following pasture. There were no differences in the belly line scores ( $2.0 \pm 0$  vs.  $2.3 \pm 0.5$ ). There was a poor correlation between the change in the sum of scores for BHR and the change in BW before and after pasture (Figure 2A).

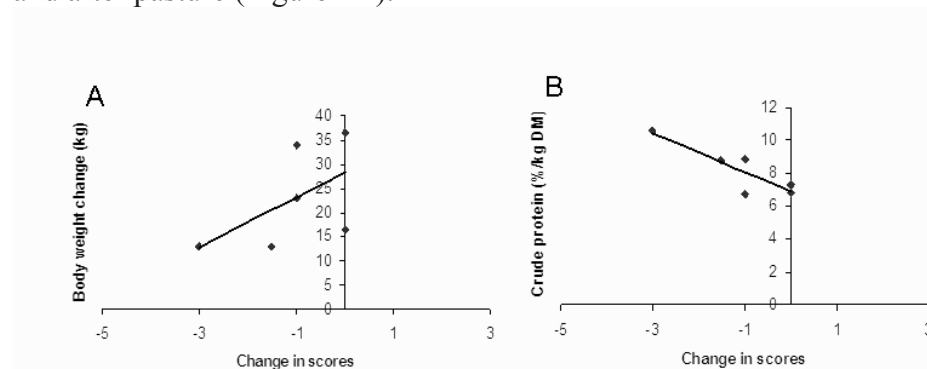


Figure 2. The correlation between changes in body condition scores (sum of back, hindquarters and ribs, BHR) and changes in body weight (A) ( $y = 5.1x + 28.2$ ,  $R^2 = 0.30$ ) in six fit Standardbred geldings before and after 31 days on pasture, and the correlation between changes in body condition scores (BHR) and faecal crude protein at the last day on pasture (B) ( $y = -1.2x + 6.9$ ,  $R^2 = 0.76$ ).

There was a significant reduction in the faecal CP content following pasture compared to the stable diet ( $9.6 \pm 1.0$  vs.  $8.2 \pm 1.5\%$  of DM). There was a correlation between faecal CP content and the loss of body scores, where the horses with lowest body score losses showed the lowest CP content (Figure 2B).

## Discussion

All horses increased their BW following pasture but no horse increased its body scores for BHR, and four of the horses had lower BHR scores. Because of the lack of increase in BHR scores (i.e. body tissue) the increase in BW was probably mainly due to increased gut fill. An increase in the score for belly was also observed in the two horses with the greatest BW increase (36.5 and 38 kg, 8.4 and 5.8%, respectively). In the other horses, the increase in BW was 2.5-4.5%, corresponding to a weight of 13-23 kg. The lack of correlation between body scores and BW changes indicates that BW change might be a poor indicator of body condition at natural pasture.

The mean CP content of the faeces was reduced following pasture and was also lower than the 8.5% that has been estimated by NRC (1978) to correspond to an intake of the maintenance CP requirement. A CP intake below requirement might explain why there was a reduction in BHR score. However, on an individual basis the horses with the lowest faecal CP maintained their BHR scores best, which makes the results difficult to interpret. There might have been individual differences in the selection of plants, their protein quality and the uptake of amino acids. It was also interesting to note that the most dominant horse had the highest faecal CP content but the greatest reduction in BHR score. A correlation between faecal CP and rank has earlier been observed by Duncan (1992). Duncan also observed that male horses may spend less time foraging and instead spend time on competition for females. Although the horses in the present study were castrated they were frequently observed standing in a grass free corner of the range which bordered to a range where mares were foraging. The dominant gelding might therefore have been selecting more than the others while foraging, but did not spend enough time foraging to meet the requirements for maintaining body condition.

## Conclusion

This study indicates that 1) natural late-summer pasture may not be adequate to maintain body condition in fit Standardbred geldings, and 2) that BW might be a poor indicator of body condition. Therefore, access to natural late-summer pasture alone cannot generally be recommended for fit Standardbred geldings at rest. Individual body condition evaluations and supplementary feeding seem to be necessary in some individuals to maintain body condition, and thereby maybe athletic fitness.

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# Long term studies to determine management practices to enhance biodiversity within semi-natural grassland communities

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## Abstract

Restoration of semi-natural grassland habitats is an objective of many agri-environmental schemes. In 1994 replicated plots were set up to determine the impact of different hay meadow management regimes on biodiversity within improved upland pasture. The treatments included sheep grazing only ( $\pm$  lime); hay cut only ( $\pm$  lime); hay cut + aftermath grazing ( $\pm$  lime); and a control treatment of grazing + fertilizer + lime. Treatments were replicated over three blocks. Over the course of 12 years, a number of assessments were carried out to assess botanical and invertebrate diversity. More recently, nutritional analyses have been carried out on samples of resulting hay. Excluding the control treatment, *Lolium perenne* cover declined from just under 60% on all plots to just under 10% for all other treatments. Total plant species per plot was estimated as 18 in the control plots in 1999. In the hay cut + aftermath grazing treatment the number of plant species is increasing each year and currently is estimated to be 70. After a 'fertility run-down phase', the hay cut + aftermath grazing treatment was the most favourable for achieving rapid diversity increases, particularly for the establishment of naturally occurring 'local' species. Nutritional analysis indicates that hay quality is within accepted nutritional standards.

Keywords: hay meadows, biodiversity, nutritional quality, grassland management, species rich grassland

## Introduction

Many agri-environment schemes provide for the restoration of semi-natural grassland habitats in the farmed landscape. However, there is a need for a greater understanding of management practices and intervention techniques to allow farmers to achieve agri-environmental outcomes whilst maintaining an acceptable level of production.

Returning agriculturally improved grassland to a semi-natural grassland community can be constrained by a lack of desirable species remaining within the seed bank, dominance of competitive species, and inappropriate grazing management. For agri-environmental management methods to be adopted by farmers the restoration methods need to be relatively non-intensive, inexpensive and practical. In addition, products that result from such management practices should be of an acceptable standard from a nutritional or quality perspective.

In recent years, there has been interest in the effects of plant species on meat and milk quality. Some studies (Fraser *et al.*, 2007; Wood *et al.*, 2007) have demonstrated increased levels of beneficial fatty acids in meat when animals grazed more species diverse pasture compared with perennial ryegrass dominated swards. There has also been interest in the nutritional quality of forage products from botanically diverse swards in terms of their trace mineral levels.

The study described within this paper is part of a Defra-funded project to compare a range of management methods for the recreation of species-rich grassland.

## Materials and methods

The experiment was set up in 1994 at the ADAS Pwllpeiran Research Centre (Lat. 52°20'N, Long. 3°50'W; elevation 310 m a.s.l.) which is situated within the Cambrian Mountains Environmentally Sensitive Area in west Wales. The site was chosen to be representative of the edaphic and climatic conditions of the region. The experiment was established on intensively managed, species-poor pasture, on an upland fringe site. Soils at the site are free-draining typical brown podzolic soils (Manod association). The mean annual rainfall is 1,770 mm. The experiment was established on a pasture which had been improved by reseeded in early the 1970s and which remained dominated by *Lolium perenne* (58% cover). Seven separate management regimes were imposed on individual plots of 0.15 ha. Treatment 1 was the control, which involved continuation of normal intensive management (i.e. limed, fertilized with NPK fertilizer and grazed by sheep; F+G+L). Treatments 2-7 represented six different options for extensification. No NPK fertilizer was applied to these treatments, which comprised a factorial designed of three cutting/grazing regimes with or without lime application. All treatments were replicated three times in a randomized block design. The '+lime' treatments received an application of lime in 1998 to raise the soil pH to approximately 6.0. Treatment 2 was a grazing-only regime with no additional lime applications (G-L); Treatment 3 was a hay cut only treatment with no lime (C-L); Treatment 4 was a combination of hay cut and aftermath grazing but no lime (C+G-L). Treatment 5 was as treatment 2 but with lime (G+L); Treatment 6 was as Treatment 3 but with lime (C+L) and Treatment 7 was as Treatment 4 but with lime application (C+G+L). For the grazing-only treatments (Treatments 1, 2 and 5) non-lactating ewes grazed the plots from April through to November each year. Where treatments included hay cuts (Treatments 3, 4, 6 and 7) these were carried out in July/August of each year depending on weather conditions. Aftermath grazing (Treatments 4 and 7) was carried out from September through to November of each year.

Changes in botanical composition were monitored annually up to 1997 and then in 2000, 2003 and 2005 by the presence and visual percentage cover estimates of vascular plants in 10 randomly located 0.4 m<sup>2</sup> quadrats per plot, during June/July. In addition, all plots were surveyed to record the presence of additional vascular species not observed within quadrats. In 2006/7, samples of hay were taken from bales from Treatments 4, 6 and 7 using a bale corer for indicative nutritional analysis.

## Results and discussion

Excluding the control treatment, *Lolium perenne* cover has declined from just under 60% on all plots to just under 10% for all other treatments. Total species per plot was estimated as 18 in the control plots in 1999. In the hay cut + aftermath grazing treatment the number of plant species is increasing each year and currently is estimated at 70. After a 'fertility run-down phase', hay cut + aftermath grazing treatment was the most favourable for achieving rapid diversity increases, particularly for the establishment of naturally occurring 'local' species. Significant increases in botanical diversity occurred in the treatments that involved both cutting and aftermath grazing. Species numbers per plot were significantly higher ( $P < 0.01$ ) in Treatments 4 and 7 (51 species) compared with the control treatment (24 species) when recorded in 2005. The increase in species was, in the main, due to an increase in desirable forb species, attaining nearly 50% cover in the most successful treatments. In the hay cut-only treatments there was also increased species numbers per plot compared with the control treatment, but this was not as great (30 and 29 species for Treatments 3 and 6, respectively). Grazing-only treatments had similar levels of increase (31 and 35 species for Treatments 2



and 5 respectively). In Treatments 2 and 5 there was also a notable increase in the undesirable species *Cirsium arvense*.

Nutritional analysis indicates that hay quality for all treatments has a high dry matter (DM) content (88.9-91.7%), and the Metabolizable Energy (ME) values and Modified Acid Detergent (MAD) fibre are acceptable. The Digestible Crude Protein (DCP) content is average, although low for the C+G-L treatment. The Crude Protein (CP) levels are slightly lower than the average for grass hay. The highest Digestibility value (D value) was for the C+G-L treatment (at 62% D) with the C+G+L treatment having a lower value of 55. The C+L treatment had a D value of 47. In consideration of the digestibility value the hay cut-only treatment has a slightly low MAD fibre value and CP, with high DM content, DCP and very high ME. The hay cut and aftermath grazing treatment is the only treatment to have a high CP and MAD fibre content. The DM and ME contents for Treatment 7 are only slightly higher than expected. Treatment 4 is the only treatment that had a hay analysis with a lower than expected ME value and DCP content.

The increase in desirable species is mainly at the expense of perennial ryegrass, which in turn affects the quality of the hay. The changing management has major implications on the agricultural value of the forages obtained. Changing management to incorporate hay cutting increased the cover of more desirable species. Plots that received an application of lime also had more desirable species as the soil had neutral pH. Hay cut-only increased the number of weedy species and therefore the hay cut and aftermath grazing was the preferred option. The late cutting date for the hay under traditional hay meadow management allowed plants to seed. These results stress the importance of maintaining neutral soil pH as this allowed desirable species-rich grasslands to develop. The botanical results also highlight the importance of the long length of time needed for restoration. After 5 years species indicative of agricultural improved grassland were still common in all plots despite the fact that a large number of semi-natural grassland species had established.

## Conclusions

The relative ineffectiveness of grazing-only treatments to increase biodiversity whilst increasing the risk of pernicious weeds limits the value of such management for agri-environment schemes. Cutting-only treatments also produced poor results. Hay cutting coupled with aftermath grazing is recommended as a management practice for the restoration of botanically diverse grassland. Preliminary investigations into the nutritional quality of forage produced from such management practices appear to be within acceptable ranges.

## Acknowledgements

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# Limited utilization impact on productivity and floristic diversity of grasslands in the Sudeten mountains

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## Abstract

The results of the research presented in this paper concern the impact of limited utilization of mountain grasslands on their productivity, species richness and floristic diversity. In 2002 an experiment was set up on an uncultivated meadow and different mowing frequencies were applied: once a year, once every two years, and once in three years, with two variants of each, the swath being removed or left. Pursuant to the results from the years 2005-2007 it was considered that favourable changes occurred in the structure and species composition of flora in the mown areas, which influenced the increase in productivity, richness and floristic diversity. The mean herbage yield ( $3.7 \text{ t DM ha}^{-1} \text{ y}^{-1}$ , across all mown treatments over 3 years) was higher by about  $0.5 \text{ t DM ha}^{-1}$  than the yield from the control area. Statistical analysis did not reveal significant differences between the level of yield from the areas of diversified mowing frequency and in variants with biomass being removed and left. The areas mown once a year and once every two years were characterized by higher number of species and higher biodiversity index. Leaving the biomass had a significant impact with a decrease in both values.

Keywords: Sudeten, grasslands, limited utilization, productivity, floristic diversity

## Introduction

Grasslands constitute natural resources of the Sudeten region due to their multifaceted importance (economic, ecological and protective) and the area they cover (120,000 ha – 40% of croplands). In the last decade of the 20th century, during the political transformations in Poland, the profitability of animal production drastically decreased, which led to a discontinuation of the utilization of grasslands and consequently to their degradation. Since 2004, after Poland's accession to the EU, farmers have made use of the financial support aimed at restoring grassland utilization. An active form of protection includes their limited utilization, i.e. cutting at least once during the vegetation season. The necessity of removing the cut biomass arouses controversy among scientists as it can lead to excessive impoverishment of the habitat.

The aim of the presented research is to specify the impact of diversified intensity of limited utilization of the mountain meadow (as required by the CAP) on the productivity, richness and floristic diversity of the sward.

## Materials and methods

This experiment was set up in 2002 on a mountain meadow in the Sudetes located 700 m a.s.l. and which had been uncultivated for 15 years. The species composition of the meadow (17 species of dicotyledons, including two papilionaceous species and 10 grass species) was similar to anthropogenic communities of meadows and pastures of *Arrhenatheretalia* (Pawl). The vast majority of species included mainly grasses: *Dactylis glomerata* L., *Phleum pratense* L., *Alopecurus pratensis* L., *Festuca pratensis* Huds., *Deschampsia caespitosa* Horv., *Poa pratensis* L., *Agrostis gigantea* Roth. and *Holcus mollis* L., and dicotyledons: *Stellaria gramineae* L., *Galeopsis tetrahit* L., *Galium mollugo* Mill. and *Veronica chamaedrys* L. The

sward was characterized by noticeable patches of the above-mentioned species. The experiment was set up with a randomized block design in four replications (plot size of 5 m x 10 m) with the following treatment areas: mown once a year, once every two years, and once every three years, with two variants, the biomass being removed (I) or left (II), and a control. Cutting was done in the first half of July. The amount of green matter from the area of 1 m<sup>2</sup> and the amount of dry matter from 1 ha was specified in the areas of each repetition. Phytosociological relevés were taken with the use of the Braun-Blanquet method and the Shannon-Wiener's diversity index was calculated. Statistical analysis of the results was carried out with the use of analysis of variance, applying F-tests. The work shows the experimental results obtained in the years 2005-2007.

## Results and discussion

In the year in which the research commenced (2002) the meadow where the experiment was set up was characterized by patches and a mosaic structure of vegetation. The introduction of cutting resulted in breaking the succession and the beginning of quantitative and spatial changes. The number of plant species in the entire experiment increased from 27 in 2002, through 30 in 2005, to 37 in 2007. Their spatial distribution and mutual relations have also changed. In the area which was cut once a year (in both variants) the structure of the sward became more heterogeneous as early as 2005. This was also noticed in the last year of research in the area cut once every two years. Phytosociological relevés revealed the increase in the quantity of medium grasses (especially in the area cut once a year) and papilionaceous plants. The smallest changes occurred in the area cut once every 3 years. The restructuring of the sward influenced its productivity as well as richness and diversity.

The areas which were cut gave a mean herbage yield across all years and treatments of 3.70 t DM ha<sup>-1</sup>, which was > 0.5 t DM ha<sup>-1</sup> more than the control areas (Table 1). The highest yield was produced in the area cut once every 3 years (3.91 t DM ha<sup>-1</sup>), and the lowest was from the area cut every year (3.50 t DM ha<sup>-1</sup>), which could be related to the increase in the amount of medium grasses which have lower yields. Regardless of the mowing frequency, the areas where the biomass was removed suggested higher yields in spite of the fact that the biomass which is left enriches the substrate with nitrogen (Barabasz-Krasny, 2002). However, statistical analysis did not confirm the significance of differences in the level of yield, either in the case of mowing frequency or the variants with the swath being removed or left.

The average number of species (Table 2) in the areas which were cut amounted to 17.43 (compared with the control, 15.17). The lowest number was recorded in the area cut once every three years (16.04), and the highest in the area cut once every 2 years (18.46). In these areas the number of species increased every year. The areas where the biomass was left were characterized by being less species-rich, which is also emphasized by other authors (Bryan *et al.*, 1998). Statistical analysis revealed the significance of differences in the number of species in the years, and in relation to mowing frequency and its variants of with or without biomass removal.

The value of the Shannon index of floristic diversity (Table 3), as well as the species diversity, was higher in each subsequent year of the research. The average value of this index in the areas which were cut was 1.82 (the control area was 1.56). The highest values were obtained in the areas cut once a year and once every two years (1.84 and 1.86), the lowest, in the area cut once every three years 1.76. In the areas with the biomass left the average values of the index were significantly lower and this concerns the areas cut once every 2 and 3 years. The area cut once a year in both variants obtained comparable values.

Table 1. Mean yields of herbage dry matter (t ha<sup>-1</sup>), 2005-2007.

		Frequency of mowing						
Years		Once a year biomass		Once in 2 yrs. biomass		Once in 3 yrs. biomass		
variants I,II	Control	I-removed	II-left	I-removed	II-left	I-removed	II-left	Significance <sup>a</sup>
2005	3.37	4.10	3.85	4.52	4.00	4.45	4.06	
2006	3.41	3.75	3.63	4.10	3.62	4.58	4.16	
2007	2.68	2.72	2.94	2.83	3.12	3.25	3.00	
Mean	3.15	3.52	3.47	3.82	3.58	4.09	3.74	
Mean frequency of mowing		3.50		3.70		3.91		ns
Mean variants I, II		I – 3.81		II – 3.60				ns

<sup>a</sup>*P* > 0.05 ns-no significance.

Table 2. Mean amount of species, 2005-2007.

		Frequency of mowing						
		Once a year		Once in 2 yrs.		Once in 3 yrs.		
Years		biomass		Biomass		biomass		
variants I,II	Control	I-removed	II-left	I-removed	II-left	I-removed	II-left	Significance <sup>a</sup>
2005	14.25	15.25	14.00	17.25	15.75	15.75	15.00	
2006	16.50	17.75	17.25	19.75	18.25	16.25	15.50	
2007	14.75	22.75	19.75	21.00	18.75	19.25	14.50	
Mean	15.17	18.58	17.00	19.33	17.58	17.08	15.00	
Mean frequency of mowing		17.79		18.46		16.04		***
Mean variants I, II		I – 18.33		II – 16.53				***

<sup>a</sup>*P* < 0.001\*\*\*.

Table 3. Mean values of Shannon-Wiener's diversity index, 2005-2007.

		Frequency of mowing						
		Once a year		Once in 2 yrs.		Once in 3 yrs.		
Years		biomass		biomass		biomass		
variants I,II	Control	I-removed	II-left	I-removed	II-left	I-removed	II-left	Significance <sup>a</sup>
2005	1.49	1.75	1.86	1.81	1.44	1.70	1.65	
2006	1.58	1.83	1.71	2.01	1.85	1.85	1.62	
2007	1.61	1.90	1.98	2.13	1.95	1.97	1.78	
Mean	1.56	1.83	1.85	1.98	1.75	1.84	1.68	
Mean frequency of mowing		1.84		1.86		1.76		ns
Mean variants I, II		I – 1.88		II – 1.76				**

<sup>a</sup>*P* > 0.05 ns – no significance; *P* < 0.01\*\*.

## Conclusions

Restoring the utilization of uncultivated mountain grasslands results in advantageous changes, both in quantity of herbage harvested, and in terms of their botanical quality. These are important for the natural and economic aspects of grassland communities. The non-removal of the biomass is the factor hindering the occurrence of these changes.

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# Crop and ley-rotation challenges for low input dairy farming systems – a planned farm study

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## Abstract

A public pilot farm (630 ha) located in Friuli Venezia Giulia region (Italy) is managed in order to promote Low Input Farming Systems (LIFS). The present High Input Farming System (HIFS) is in transition to LIFS using Good Farming Practices to qualify for the agri-environment. Seeds suitable for LIFS will be tested; techniques of soil management such as minimum tillage and direct drilling will be used. Traditional rotation maize-soybean will be improved with more environmentally friendly crop rotation systems introducing minor cereals, legumes and multi-species meadows for ley-arable rotation. 258 ha will be used for experimental trials to improve soil fertility and carbon sequestration, to reduce fertilizing intensity, to protect soil from erosion, to maintain soil organic matter and soil structure and to reduce the use of pesticides. Crop rotation, soil cover, fertilizing intensity, fertilizer uptake, nitrate leaching, manuring, and landscape elements will be used as indicators.

Keywords: crop rotation, low-input farming systems, energy balance, dairy cows

## Introduction

Human activity has contributed to create and maintain a variety of semi-natural habitats, but high input farming systems (HIFS) have had a profound impact on nature conservation and landscape integrity throughout much of Europe. In areas where intensive livestock and agronomic production is practised a great loss of fauna and flora has been registered.

Recently the EC defined rules and regulations in order to improve agricultural practices in terms of preserving the environment, and to help farmers meet the standards of high quality agriculture and new society's expectations. Public experimental and demonstration farms (PEDF) can be good examples to promote environmentally friendly agriculture and to communicate the results to policymakers and the wider public. In PEDF the past HIFS may be converted to low input farming system (LIFS) using good farming practices (GFP) to qualify for the agri-environment, and to drive farmers to a new model of agriculture.

The aim of the study is to present the agronomic resettling programme of a public pilot farm located in the Friuli Venezia Giulia region (Italy). It was cultivated until 2005 using HIFS, but a transition period and a requalification programme started in 2006 and will be concluded in 2012 to reduce significantly external inputs, to adopt environmentally friendly techniques, i.e., by more ley-arable rotations, and to introduce landscape elements.

## Materials and methods

The Marianis-Volpares farm (630 ha) is owned by the Regional Government of Friuli Venezia Giulia and is managed by the Regional Agency for Rural Development (ERSA). The farm is located in the municipality of Palazzolo dello Stella (Udine, Italy, Lat. 45°48'0"N, Long. 13°5'0"E, Alt. 0-8 m a.s.l.).

The soil is clay-loamy (pH 7.8-8.0). Irrigation is provided on maize and soybean. There are 529 ha of cultivated land. The main crops are maize (174 ha), soybean (136 ha), lucerne and grassland (109 ha), barley and wheat (39 ha), poplar plantation (22 ha), rape (1 ha), sunflower



(5 ha), and set aside (43 ha). The Marianis-Volpares farm was cultivated until 2005 by HIFS and crops (maize, soybean and poplar) cultivated for selling the products. The present total amount of fertilizers used on the farm is about 7,000 m<sup>3</sup> y<sup>-1</sup> of dung from the farm, and 33 Mg y<sup>-1</sup> of N, 18 Mg y<sup>-1</sup> of P and 10 Mg y<sup>-1</sup> of K from mineral fertilizers. An important reduction of fertilizers and input of energy from outside (-0.2) started in the year 2006. It will be much more significant in the next experimental and transition period (2008-2012).

A large part of the farm (258 ha) will be employed for experimental trials to train farmers and advisory officers. The main rotational plans will be improved using more ley-arable land, i.e. (A) 1-year rotation maize silage+ryegrass; (B) 2-crop rotations i.e. (B1) maize silage/sorghum-barley+soybean; (B2) ryegrass+sorghum-grain pea+foxtail millet; (B3) maize+pea/landsberger-sorghum+rape/crimson clover; (B4) rape-triticale/ rye/ ryegrass/ barley+soybean; (B5) maize-soybean; (C) 3-crop rotations i.e. (C1) silomaize-ryegrass + soybean-wheat/crimson clover/fodder vetch; (C2) sunflower-wheat-rape+foxtail millet; (C3) silomaize-ryegrass+soybean-triticale/silorye+sorghum/foxtail millet; (C4) maize+horse radish-soybean-wheat+crimson clover/fodder vetch; (D) 4-crop rotations i.e. (D1) sunflower+crimson clover-maize+crimson clover+ryegrass-soybean-wheat; (D2) rape+soybean-maize+crimson clover-soybean-wheat; (E) 5-crop rotations or more i.e. (E1) triticale/silo rye+soybean-maize-sunflower+green manure pea-silo maize-ryegrass+soybean; (E2) maize-barley+lucerne-lucerne-lucerne-lucerne; (E3) maize-barley+soybean-wheat+lucerne-lucerne-lucerne-lucerne; (E4) maize-barley+soybean-silomaize-lucerne-lucerne-lucerne. Yields will be used almost totally to feed animals.

## Results and discussion

The main agronomic treatments used for the crop rotations and expected yields (dry matter, crude protein, milk feed units) according to previous experiments, are summarized in Table 1.

Table 1. Main agronomic treatments for the crop rotations and expected yields according to previous experiments.

COD	Surface (ha)	Dry Matter (Mg/ha/y)	Crude Protein (Mg/ha/y)	Milk Feed Unit (Mg/ha/y)	Slurry	Manure	Years	Cover crops	Minimum tillage
A1	10	26.3	2.2	21,000	yes	no	1	no	no
A2	10	17.5	1.5	15,000	yes	no	1	yes	yes
B1	10	12.8	1.6	12,200	yes	yes	2	no	yes
B2	10	17.3	1.7	13,900	yes	no	2	yes	yes
B3	10	8.3	0.9	10,150	yes	no	2	yes	no
B4	10	6.7	1.0	5,600	yes	yes	2	yes	yes
B5	30	6.1	1.2	7,750	yes	yes	2	no	no
C1	12	9.8	1.2	8,450	yes	yes	3	yes	yes
C2	12	2.9	0.2	2,000	yes	yes	3	no	yes
C3	12	16.0	1.6	13,200	yes	yes	3	no	yes
C4	12	4.1	0.8	5,100	yes	yes	3	yes	yes
D1	16	3.1	0.6	3,850	yes	yes	4	yes	yes
D2	16	3.6	0.8	4,550	yes	yes	4	yes	yes
E1	20	10.4	1.3	9,550	yes	yes	5	yes	yes
E2	20	8.3	1.1	6,750	yes	yes	5	no	yes
E3	24	7.4	1.1	6,130	yes	yes	6	no	yes
E4	24	9.2	1.2	7,950	yes	yes	6	no	yes

Maize and soybean are the most important crops in this area for economic and agronomic reasons and for animal feed. Proportionately, about 0.6 of the maize will be cultivated for silage production and 0.3 for maize grain production to be employed on the farm, while 0.1 will be used to produce biomass to feed a digester to produce biogas. Earlier cultivars for



grain maize (400-500 FAO) and for silage maize (600 FAO) will be employed in order to reduce pests, diseases (e.g. corn borer, western corn root worm, etc.) and drying energy. Cultivars with a deep root system and prostrate leaves will be chosen over more popular varieties with erect leaves and small root systems, which are highly productive but require much more water and fertilizer. Maize will be never cultivated in monoculture. Soybean varieties selected by ERSA with low anti-nutritional factors content (trypsin inhibitor, lipoxigenase activities), will be cultivated, dried and employed to feed animals directly on the farm. A larger area will be cultivated with lucerne to improve N in crop rotation systems (Lyons and Latta, 2003), and arable land will be converted to multi-species grassland in order to improve hay in the animal diet (Wachendorf *et al.*, 2005). The past monoculture system of maize or traditional rotation maize-soybean and lucerne will be integrated with more environmentally friendly crop rotation systems introducing minor cereals (e.g. sorghum, wheat, barley) and legumes (e.g. pea). In order to save water and to anticipate possible problems of western corn root worm a limited area with grain sorghum and grass sorghum will be introduced. In fact, this crop is less water demanding and more pest resistant. Legume winter and summer cover crops (red clover, crimson clover, hairy vetch etc.) will be used for 'green manuring' as well as incorporated into the soil (Schroeder, 2000). Grasses like ryegrass will be established after the main crop (e.g. maize or soybean) and used primarily to reduce nutrient leaching from the soil profile and obtain important biomass (silage or hay) for animal feeding. Barley and wheat, already used, will be also improved both for providing more biomass for livestock and to be used as a catch crop. Organic fertilizers produced on the farm, after digestion processes to reduce possible pollution, will be employed in the crop rotation systems. Chemical fertilizers will be used only in quantities which respect the ratio out/input in the plant/soil system in order to save energy and avoid environmental problems. Crop rotation will also be used in order to improve soil fertility and carbon sequestration, to reduce fertilizing intensity, to protect soil from erosion, to maintain soil organic matter and soil structure, and to reduce the use of pesticides. Crop rotation combined with improved tillage practices may have an important role in limiting water and wind erosion.

## Conclusions

The period of conversion from HIFS to LIFS, during which lower yields and increase of investments with or without the benefit of premium prices will occur, will be certainly very difficult financially and therefore farmers must be guided through it. Experimental and demonstrative farms like Marianis-Volpares may be important tools to meet at least some of the most important agricultural and environmental policy objectives.

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# **Vegetation type selected by cattle grazing heterogeneous semi-natural pastures**

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## **Abstract**

Three 10-15 ha paddocks with heterogeneous semi-natural pasture, grazed by Hereford suckler cows with calves or Swedish Friesian and Swedish Red, were used in the study. In each paddock, sub-areas with different vegetation types were identified and mapped: dry, mesic, wet, wooded and previously cultivated. In each of the five vegetation types in all three paddocks, three 1 x 1 m plots were established where nutrient content and botanical composition were analysed. The time the animals spent grazing on the different vegetation types was studied. Registrations of the vegetation type that cattle selected were performed every five minutes during four grazing hours, three consecutive days in June, July and August. The animal behaviour data was analysed in a statistical model that included vegetation type and the proportion of pasture area each vegetation type represented. Neither the effect of paddock nor of month was significant.

The highest nutrient content (energy and crude protein) of the available herbage could be found in the previously cultivated areas, and the results show that the animals chose to graze previously cultivated vegetation on 68% of the total 990 observations.

Keywords: grazing, semi-natural pasture, selection, vegetation type

## **Introduction**

Do cattle grazing semi-natural pastures prefer certain types of vegetation? What is the nutrient content of different vegetation types in semi-natural pastures and is there a connection between a crop's nutrient content and the cattle's selection?

An animal's selective behaviour varies with grazing pressure, i.e. how many animals there are per unit of available forage. The more forage an animal has access to, the more selective it can be. When given a choice, animals have been shown to prefer forage with high nitrogen and energy content but low fibre content (Arnold, 1981; Pehrson, 2001). However, since Swedish semi-natural pastures are very heterogeneous in nature, there are few comprehensive studies of the nutrient content in such pastures.

The object of this study was to ascertain whether cattle grazing semi-natural pastures prefer certain types of vegetation, and if so, if this preference is related to the nutrient content of the forage.

## **Materials and methods**

The experiment was carried out during June, July and August 2006, in three different semi-natural pastures of approximately 10-15 ha each, with stocking rates of 0.44, 0.89 and 4.96 animals ha<sup>-1</sup>. The paddocks were chosen for their heterogeneity, so that the selective behaviour of the cattle could properly be observed. Each paddock was then mapped and divided into sub-areas where vegetation type – dry, mesic, wet, wooded, or previously cultivated – was noted. Two of the paddocks were grazed by Hereford suckler cows with calves, while the third paddock was grazed by milk cows of the Swedish Friesian and Swedish Red breeds.

A behaviour study was carried out in which the animals (the calves and bulls not included) were observed during four hours of active grazing each day, three consecutive days per month. Every five minutes, the area and type of vegetation the animals grazed was noted. In addition, a sample of the vegetation grazed by the animals was taken for analysis. These smaller samples were pooled into one 'day-sample'.

Three 1 x 1 m sample plots were laid out in each of the five vegetation types, thus making fifteen sample plots in each of the three paddocks. Once every month, typically the day following the three behavioural study days, the vegetation in the plots was cut. The samples were then prepared and analysed with regard to dry matter (DM), ash and crude protein (CP) (Kungliga Lantbruksstyrelsens Kungörelser nr 15, 1966) and neutral detergent fibre (NDF) (Van Soest and Wine, 1967). The content of metabolizable energy (ME) was determined by *in-vitro* digestion as described by Lindgren (1983). The same analyses were carried out on the vegetation samples taken during the behavioural study.

The statistical analysis was made in SAS 9.1. Three statistical models were used in the experiment: one to examine the nutritive content of the vegetation types, one to determine which variables had an influence on the available amount of forage in different areas at the time of observation, and one to determine which variables contributed most to the animal's choice of grazing area. As animal breed differences were not the object of this study this variable was not therefore included in any model.

## Results and discussion

In all three paddocks, the animals preferred to graze the previously cultivated vegetation type and spent 68% of the total observed grazing time on these areas over the season. One may assume that the animals would graze each area according to its relative size. However, when comparing the size of each vegetation type with the amount of time grazed there, the time the animals grazed on the previously cultivated vegetation was higher than expected (Figure 1).

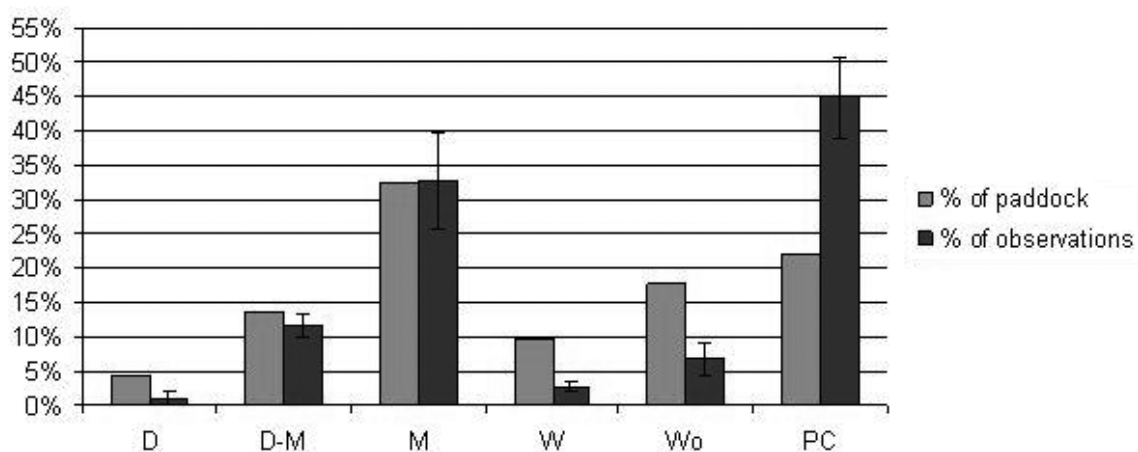


Figure 1. Proportion of the time that animals spent grazing on different vegetation types (averaged over entire season) in relation to the size of that vegetation type. The figure contains raw data from paddock 3. D = dry, D-M = dry to mesic, M = mesic, W = wet, Wo = wooded, PC = previously cultivated.

Statistical analysis of the animals' choice of grazing area showed that vegetation type ( $P < 0.001$ ) was highly significant as a class variable together with the proportion of paddock area each vegetation type represented. The content of CP ( $P < 0.001$ ) was also significant but did not increase the likelihood of the model that contained vegetation type as a class variable. The quantity of forage available in the different vegetation types at the time of observation did not

influence the grazing time on each vegetation type. The analysis of the sample plots showed that the wet vegetation type had the most available forage at the time of observation, and that the wooded type tended to have the least. It should be noted that there was no study made of the overall productivity of the vegetation types in this study, only of the amount of available forage at specific times.

Previously cultivated vegetation had a higher CP content than the other vegetation types ( $16.2 \text{ g kg}^{-1} \text{ DM}$ ) whereas the lowest CP content could be found in the wooded, dry and wet areas ( $10.4$ ,  $10.8$  and  $11.4 \text{ g kg}^{-1} \text{ DM}$ , respectively). There was no interaction between paddock and vegetation type. The analysis of factors that affected the content of ME and NDF in the vegetation samples showed an interaction between paddock and vegetation type (i.e., the energy and fibre content in different vegetation types varied between paddocks). However, the highest energy contents in all three paddocks were found in previously cultivated areas, ranging between  $9.7$  and  $10.1 \text{ MJ kg}^{-1} \text{ DM}$ , whereas the lowest contents were found in the wet areas with ME values ranging between  $7.5$  and  $8.6 \text{ MJ kg}^{-1} \text{ DM}$ . In a similar manner, the previously cultivated areas had lower NDF contents ( $402\text{--}524 \text{ g kg}^{-1} \text{ DM}$ ) than areas of wet vegetation ( $533\text{--}606 \text{ g kg}^{-1} \text{ DM}$ ). More detailed information about CP, energy and NDF contents in the different vegetation types can be found in Pelve (2007).

Because the animals spent 68% of their available time in areas of the previously cultivated type, which had the highest energy and protein content and the lowest NDF content in most cases, one can assume that the animals choose this vegetation according to its nutrient value.

## Conclusions

Vegetation type explained the animals' selective behaviour better than any quality variable by itself. The classification of vegetation into dry, mesic, wet, wooded and previously cultivated areas is therefore relevant to understand and predict grazing behaviour. The nutrient content of different types of vegetation varied substantially within the paddocks, from the wet high-fibre, low-energy type to the previously cultivated high-energy, low-fibre type. In this study, the animals chose to graze in areas where the vegetation had the highest nutrient content (previously cultivated vegetation) rather than in areas with the highest quantity of available forage. However, the selective behaviour might have been different if grazing pressure had been higher.

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## Effect of shading on biodiversity of cocksfoot (*Dactylis Glomerata*) established sward

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### Abstract

Silvopastoral systems are sustainable systems of land management as defined in the Agenda 21. The presence of trees in agroforestry systems reduces the quantity of light that reaches the understorey compared with open swards. The different inputs of radiation can cause modifications in the competition relationships of the pasture plants. This experiment aims to evaluate the shading effect on the temporal evolution of different herbaceous species after establishment with *Dactylis glomerata* L., cv. Artabro. The experiment was carried out in Lugo (NW Spain, 456 m. above sea level) in 1996 (after sowing with 30 kg ha<sup>-1</sup> of cocksfoot) and in 1997. Two treatments were established: a) open pasture and, b) shading, which was simulated with a plastic mesh that intercepted 50% of incident radiation that reaches the pasture. Species richness was greater after sowing of cocksfoot. Shading positively affected species richness during the spring, but it negatively affected species richness in the autumn.

### Introduction

In Galicia (NW Spain) around 62% of the area is occupied by forests, largely created due to afforestation subsidies from the government and based on the European Union Policies. Approximately 15% of the Galician area has been afforested in the last ten years (Rigueiro-Rodríguez *et al.*, 2005).

Silvopastoral systems are being promoted by the autonomous administration in Galicia, because they enhance forest production in both the short and the long term, and because there is a high degree of forest fires in the area (Rigueiro-Rodríguez *et al.*, 2006). In addition, biodiversity can be promoted by silvopastoral systems because they generate different light, temperature and humidity gradients under the trees, with different microclimatic conditions (Herzog, 2000). Radiation input which reaches the understorey can modify botanical composition (Silva-Pando, 1988). Increased knowledge about effects of shading on species biodiversity will allow predictions on the impact of this land-use management on biodiversity (Rigueiro-Rodríguez *et al.*, 2007).

### Materials and methods

The experiment was carried out in Lugo (NW Spain, at an altitude of 456 m a.s.l.) in 1996 and 1997. Pasture was established in six plots in autumn, after sowing with 30 kg ha<sup>-1</sup> of cocksfoot (*Dactylis glomerata*) in plots of 1 m<sup>2</sup>. A randomized block design with three replicates was used, with two treatments: open habitat and artificial shading. Artificial shading was simulated with a plastic mesh that intercepted 50% of incident radiation that reaches the pasture.

Pasture was sampled in April, June, July, October and November in 1997. The October sampling was made due to the unusual rainfall during the summer. Sampling consisted of harvesting two squares of 0.3 x 0.3 m in each experimental unit at a height of 2.5 cm above ground level. Samples were transported to the laboratory, where botanical composition was estimated by hand separation and determined to species level. Samples were weighed after drying in an oven (48 h at 60 °C). Treatments were evaluated with ANOVA using the statistics program SAS (2001), and significance of means determined using LSD test.



## Results and discussion

Different species were found in the different harvests (Table 1). The sown species *Dactylis glomerata* was found in both treatments and during all the year. Species like *Plantago lanceolata* L. and *Holcus lanatus* L. were only present in the shaded treatment and other species like *Taraxacum officinale* Weber, *Trifolium* spp. and *Rumex obtusifolius* L appeared in both treatments during the whole year. Species richness (Figure 1) was greater in herbage at the first than in the last harvest of the year ( $P < 0.05$ ). This could be explained by the presence of gaps during the first part of the year after sowing, which would allow pasture species to grow without competition. In the middle of the spring, light conditions enhanced productivity which is associated to a higher pasture height, and which significantly reduced pasture richness. At the end of the spring, height of pasture was probably high in both open and shaded plots. Tall vegetation leads to patches without vegetation in the top-soil, being most important in light conditions (mid-spring) which enhanced biodiversity at the end of the year (Mosquera-Losada *et al.*, 2004). This is very important when tree cover is not complete, as pasture species development is different under the tree than in open pasture. The heterogeneity generated by the tree enhances biodiversity (Rigueiro-Rodríguez *et al.*, 2007).

Table 1. Pasture species presented under different analysed treatments (shadow and light) in the different harvests in 1997.

	Apr 1997		Jun 1997		Jul 1997		Oct 1997		Nov 1997	
	Shadow	Light	Shadow	Light	Shadow	Light	Shadow	Light	Shadow	Light
<i>Trifolium pratense</i>	*	*	*	*	*	*	*	*	*	*
<i>Trifolium repens</i>	*	*	*	*	*	*	*	*	*	*
<i>Rumex obtusifolius</i>	*	*	*	*	*	*	*	*	*	*
<i>Capsella burspastoris</i>	*	*				*				
<i>Sonchus oleaceus</i>			*	*	*	*				
<i>Veronica persica</i>	*	*			*	*				*
<i>Taraxacum officinale</i>	*	*	*	*	*	*	*	*	*	*
<i>Lolium perenne</i>				*						
<i>Lolium multiflorum</i>			*	*		*		*		
<i>Stellaria media</i>	*	*			*	*	*		*	*
<i>Dactylis glomerata</i>	*	*	*	*	*	*	*	*	*	*
<i>Erodium moschatum</i>	*	*		*		*		*		*
<i>Plantago lanceolata</i>	*		*							
<i>Solanum nigrum</i>							*			
<i>Senecio vulgaris</i>										*
<i>Festuca arundinacea</i>	*									
<i>Holcus lanatus</i>	*		*							
<i>Cerastium glomeratum</i>	*	*			*					
<i>Lamium purpureum</i>	*	*								*
<i>Amaranthus retroflexus</i>										*
<i>Crepis capillaris</i>			*		*					
<i>Poa annua</i>	*	*	*		*	*				
<i>Lotus uliginosus</i>										
<i>Epilobium tetragonum</i>	*	*								
<i>Coleostephus myconis</i>					*					
Tot	16	13	11	9	12	12	7	7	6	11



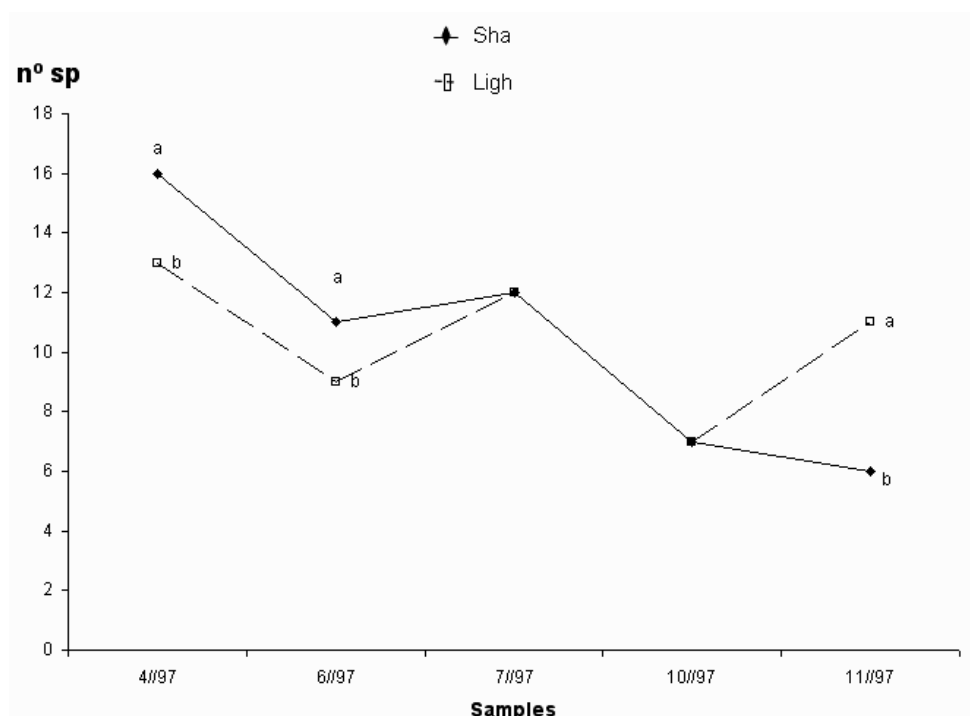


Figure 1. Richness (number of species) under both treatments in April-November 1997.

## Conclusions

Pasture gaps created by the small growth during the first year, as well as the effect of tall pasture at the end of the spring, increased pasture species richness in the subsequent harvests.

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# Weight gain among steers grazing semi-natural pastures every other year compared with every year

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## Abstract

The weight gain among steers grazing semi-natural pastures every other year (Intermittent treatment) was compared with grazing every year (Continuous treatment) during 4 years. The steers were of the Swedish Red breed. The Intermittent treatment area had 2 paddocks that were alternately grazed every other year and the Continuous area consisted of only 1 paddock. Grazing pressure was monitored by sward height measurements and pasture samples were taken to evaluate the nutritive content of grazed herbage. Average sward height of the pasture on the Intermittent treatment was 1 cm higher and energy content of herbage samples 0.6 MJ per kg dry matter lower. Average daily weight gain per animal over the four years was 0.70 and 0.72 kg for the Continuous and Intermittent treatments, respectively, which did not differ significantly.

Keywords: grazing, semi-natural pastures, cattle, management

## Introduction

Semi-natural pastures in Sweden have large biodiversity values, but there is a concern that many of these pastures will be abandoned in the future due to a shortage of grazing animals. Grazing every other year would help preserve semi-natural pastures with fewer animals. However, it is possible that large amounts of wilted and senescent herbage would accumulate if the pasture is left ungrazed over an entire season, with negative effects on botanical biodiversity and animal weight gain during the following grazing season. Wissman (2006) found the effects of grazing every other year to be favourable for biodiversity. This paper focuses on evaluating the effect of grazing every other year on the weight gain of steers.

The weight gain of the animals on the Intermittent treatment was expected to be lower compared with the animals on the Continuous treatment due to large amounts of low-quality senescent herbage from the ungrazed vegetation left from the previous season in the Intermittent pasture. The objective of the experiment was to evaluate the effects on animal weight gain, of grazing every other year compared with grazing every year.

## Materials and methods

Three low-productive semi-natural pastures were used in the study. All three were located on a farm (at 59°6'N, 16°29'E) where the grazing season lasts from early May until early October. The semi-natural pasture areas used in the experiment all had a long history of yearly continuous grazing when the experiment started in 2001, and they were similar in vegetation structure but differed in size. Two pastures were used for the Intermittent treatment group and were alternately grazed every other year, and thus alternately left ungrazed the year in-between. The pasture used for the Continuous treatment was continuously grazed every year. Weight gain measurements were taken during four years, starting in May 2002 when the animals on the Intermittent treatment started to graze the pasture that had been left ungrazed the previous season. The results are based on a total of 57 steers of the Swedish Red breed

with 38 and 19 animals on the intermittent and continuous treatments, respectively. Average live weight (standard deviation SD) at the start of the grazing season was 251 (SD 44) and 240 (SD 51) for the intermittent and continuous treatment, respectively. All animals were treated with anthelmintics before the start of the grazing season for protection against gastrointestinal nematodes. As the grazing pressure was regarded to be somewhat high during the first two years, the number of animals were reduced by 25% during the last two years of the experiment (2004-2005) from 1 to 0.75 animals per ha.

Grazing pressure was monitored by sward height measurements using a 30 x 30 cm, 430 g falling plate meter. Measurements were performed every second week along a number of defined transects, with at least 60 measurements per treatment and measuring occasion in 2002-2004 and double that in 2005. The plan was to adjust the number of grazing animals by removing or adding non-experimental grazers when the sward height differed more than 1 cm between treatments on two successive measuring occasions. Herbage sampling was also performed along the same transects at 30 points. Sward samples used for the analysis of herbage nutritive value were plucked by hand to simulate cattle 'bites'. In 2005, herbage samples were collected by following the animals in each treatment group for 1-2 hours of grazing and sampling close to the herbage eaten. Herbage samples were analysed with regard to dry matter (DM), crude protein (CP) and Neutral Detergent Fibre (NDF), according to conventional procedures. The content of metabolizable energy (ME) was determined by *in-vitro* digestion as described by Lindgren (1979).

Animals were weighed four times: in February, early May (at pasture let-out), July and September/October. The weight gain between February and early May, when animals from both treatments were together in the barn and on the same diet, was used as a covariate in the statistical analysis of the weight gain during the grazing season. Analysis of variance was performed using the GLM procedure in SAS (2002). Animal weight gain during 2002-2005 was analysed in a statistical model containing treatment, stocking rate and year nested within stocking rate. Weight gain of the steers indoors before start of the experiment was treated as a covariate in the model. As the effects of year nested within stocking rate and the interactions between treatment, stocking rate and year nested within stocking rate were not significant, these variables were removed from the model. The final model contained only the effect of treatment (Intermittent vs. Continuous) and stocking rate (high vs. low).

## Results and discussion

The average sward height on the Intermittent treatment was 6.5 cm, which was 1 cm higher than on the Continuous treatment, thus indicating a higher herbage allowance for animals on the Intermittent treatment. However, there was no statistical relationship between sward height and animal weight gain, either due to treatment, or time of year.

The average contents of ME per kg dry matter in the pasture samples from the Intermittent and Continuous treatments over the four years were 9.2 (SD 0.9) and 9.8 (SD 0.9) MJ kg<sup>-1</sup> dry matter, respectively. Although the energy contents of the herbage samples from the Intermittent pasture were lower compared with the Continuous, corresponding values for crude protein and NDF contents in the herbage samples did not differ much between treatments. Crude protein content was 154 (SD 17) and 153 (SD 21) g kg<sup>-1</sup> dry matter and NDF content was 517 (SD 49) and 494 (SD 35) g kg<sup>-1</sup> dry matter for the Intermittent and Continuous treatments respectively.

Over the four years, the weight gain of animals on the Intermittent treatment was as high as weight gain of animals in the Continuous group (Table 1). In early season, weight gain in the Intermittent group was even significantly higher than in the Continuous group.

The higher energy content in the pasture samples from the Continuous treatment was not reflected in a higher weight gain of the animals. It is probable that the pasture sampling technique during the years 2002-2004 did not reflect what the animals actually consumed. The herbage selected by the animals was monitored in more detail in 2005 and that year no differences were found in the nutritive content of the herbage selected by the animals in the two treatment groups. A more detailed presentation of this study is found in a separate paper in the proceedings of this conference (Spörndly, 2008).

Table 1. Average daily weight gain ( $\text{kg animal}^{-1} \text{ day}^{-1}$ ) at different treatments: grazing every other year (Intermittent) vs. every year (Continuous) and at different stocking rates: 1 animal  $\text{ha}^{-1}$  (High) vs. 0.75 animal  $\text{ha}^{-1}$  (Low). Least square means with standard error in parenthesis.

<i>Effect of treatment</i>	Intermittent	Continuous	<i>P</i>
No. of animals	38	19	
Early season (May-July)	0.61 (0.02)	0.52 (0.03)	0.011
Late season (July-Sept/Oct)	0.86 (0.03)	0.93 (0.04)	0.160
Early + Late Season (May-Sept/Oct)	0.72 (0.02)	0.70 (0.02)	0.411

<i>Effect of stocking rate</i>	High	Low	
No. of animals	33	24	
Early season (May-July)	0.43 (0.02)	0.69 (0.03)	< 0.0001
Late season (July-Sept/Oct)	0.85 (0.03)	0.95 (0.04)	0.07
Early + Late Season (May-Sept/Oct)	0.61 (0.02)	0.80 (0.02)	< 0.0001

The decrease in number of animals during the last two experimental years gave a higher live weight gain (approximately +0.2 kg per animal and day) over the season and in the early season. In late season there was a tendency for a higher live weight gain at the lower stocking rate. However, it must be emphasized that even though each of the stocking rates were applied during two separate years, the effect of stocking rate cannot be separated from the effect of the two experimental years when the stocking rate was applied.

## Conclusions

In this four-year experiment, steers grazing a pasture that had been left ungrazed the previous season had the same daily weight gain as steers grazed conventionally. Therefore, in similar areas where there is a shortage of animals but a need to maintain an open landscape by grazing, it is possible for these types of semi-natural pastures to be grazed every other year without leading to reduced animal weight gains

## Acknowledgements

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# Nutritive value and composition of vegetation selected by steers grazing semi-natural pastures every other year compared with every year

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## Abstract

The herbage selected by two groups of steers grazing different pastures was compared: one pasture was grazed every other year (Intermittent) and had thus been left ungrazed the previous season, while the other pasture was grazed every year (Continuous). Vegetation selected by steers in the two groups was sampled on eleven occasions between 11 May and 5 October. Each sample was sorted into three fractions: grasses/sedges, forbs/browse and senescent herbage. The nutritive contents of all samples were analysed. The results were similar for the two treatments. On a dry matter basis averaged over the season, both pastures consisted of approximately 59% grasses/sedges, 24% forbs/browse and 17% senescent herbage. The nutritive value of selected vegetation in the Intermittent and Continuous treatments, respectively, were as follows (seasonal mean, SE in brackets): 9.7 (0.3) and 9.6 (0.4) MJ per kg dry matter, 157 (7) and 146 (8) g crude protein per kg dry matter, 470 (12) and 483 (13) g neutral detergent fibre per kg dry matter. It was concluded that there were no major differences in either the composition, or the nutritive value, of herbage selected by the steers grazing the pasture that had been left ungrazed the previous season compared with herbage selected by the steers in the Continuous group.

Keywords: semi-natural pastures, cattle, nutritive content, selection

## Introduction

Semi-natural pastures that have not been subjected to N-fertilization often have a high biodiversity. However, farming is continuously being intensified and rationalized with a higher productivity per animal and it is estimated that the number of grazing animals will decrease in coming decades. Therefore, many low-productive, semi-natural pastures risk being abandoned within the next few years. This is a major concern for nature conservation because grazing is vital to many red-listed species found on these pastures. Grazing semi-natural pastures every other year would help preserve these pastures, while requiring fewer animals. The effects on biodiversity of this management practice are favourable and have been presented elsewhere (Wissman, 2006). This paper focuses on evaluating the effect of grazing every other year on the composition of feed selected by grazing animals.

The vegetation selected by animals on a pasture that had been left ungrazed the previous season (Intermittent treatment) would contain more senescent material and have a lower energy and protein content but a higher NDF content compared with the vegetation selected by animals grazing an area that was grazed each year (Continuous treatment). The difference would be largest in the early season due to large amounts of senescent herbage from the ungrazed vegetation left from the previous year in the Intermittent grazing area.

The objective was to study the composition and nutritive value of the vegetation selected by cattle on semi-natural pastures under an intermittent or continuous management strategy.



## Materials and methods

Two similar, low-productive, semi-natural pastures were used in the study. The study was performed on a farm (located at 59°6'N, 16°29'E) where the grazing season lasts from early May until early October. Both pastures had a long history of yearly continuous grazing when the experiment started in 2001. One pasture (Intermittent) was continuously grazed in 2001 and 2003 but left ungrazed and uncut throughout the 2002 and 2004 seasons. The other pasture (Continuous) was continuously grazed all four years. The stocking rate was 0.7 animals ha<sup>-1</sup> (300 kg live weight). Thirteen steers of the Swedish Red breed were observed during the grazing season 2005. At five-minute intervals, the pasture immediately adjacent to the vegetation that the animals were eating was sampled for an average sampling time of 1.7 hours (range 1-2 hours) and 20 samples per treatment and sampling occasion. These sub-samples were pooled into one sample per treatment and sampling occasion for analysis. Sampling took place between 0600 and 1800 hours and the order between paddocks varied so that animals in all paddocks were observed both mornings and afternoons. This was performed approximately every second week from 11 May to 5 October for a total of eleven sampling occasions. Herbage samples were sorted into three fractions: grasses and sedges (henceforth called grasses), forbs and browse (henceforth called forbs) and senescent herbage. Each fraction was analysed separately with regard to dry matter (DM), crude protein (CP) and Neutral Detergent Fibre (NDF) contents. The content of metabolizable energy (ME) was determined by *in-vitro* digestion as described by Lindgren (1979). Analysis of species composition in the herbage samples was performed on one occasion in August. As there is a large seasonal variation in forage composition, a paired Students t-test was performed to compare the two treatments at the different sampling occasions throughout the grazing season.

## Results and discussion

The average proportions of senescent vegetation, grasses and forbs in the vegetation selected by steers in the two treatments were similar, with a fairly constant proportion of grass in their diet throughout the season (Figure 1).

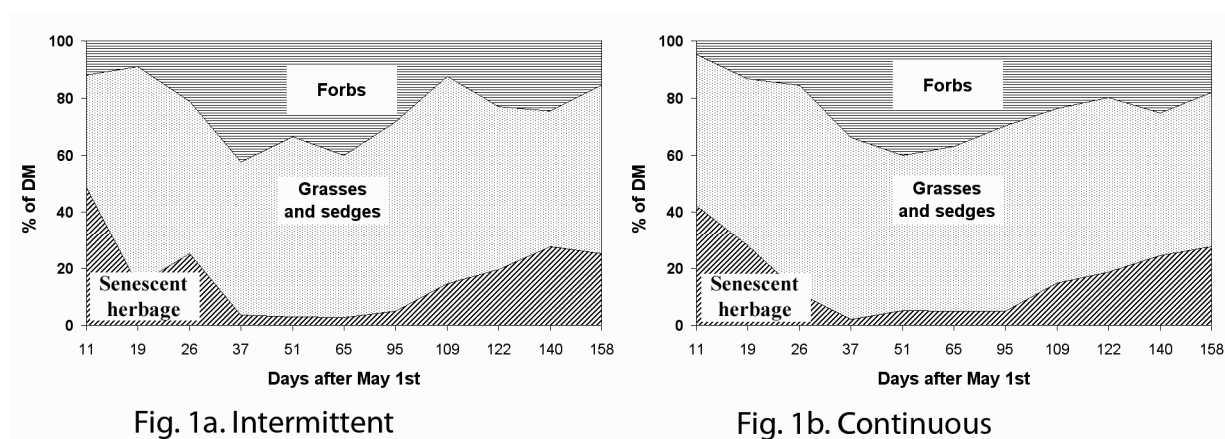


Figure 1a-b. Proportions of senescent vegetation, grasses and sedges and forbs in the vegetation selected by steers on the Intermittent (a) and Continuous (b) treatments. The figure is based on results from 11 sampling occasions.

In mid-season, the proportion of forbs was fairly high (30-40%), although it was considerably lower in early- and late-season (10-20%). The proportion of senescent vegetation showed the opposite pattern, being high early (above 40%) and late (20-30%) in the season and very low



in between (2-5%). The average proportions of senescent vegetation, grasses and forbs were similar for the two treatments in paired comparisons over sampling occasions (Table 1).

The mean CP, NDF and ME contents in vegetation samples over the entire season from the two grazing areas are presented in Table 1. After a slight initial increase, a decrease in energy content was observed as the season progressed, from 11.2 MJ kg<sup>-1</sup> DM in early June to 8.2 MJ kg<sup>-1</sup> DM in early October. Energy and NDF contents were similar between treatments, but a slightly higher CP content was observed in the herbage samples from the intermittent treatment.

Table 1. Proportions of vegetation type and the nutritive content of vegetation selected by steers grazing on a pasture that had been left ungrazed the previous year (Intermittent) or a pasture grazed every year (Continuous). Seasonal means are presented with standard error of difference (SED) and *P* value.

		Intermittent	Continuous	SED	<i>P</i>
Proportions, % of DM	Senescent vegetation	17.2	16.8	2.0	0.85
	Grasses and sedges	59.0	59.5	3.4	0.88
	Forbs and browse	23.8	23.7	1.8	0.97
Nutrient content	ME, MJ kg <sup>-1</sup> DM	9.7	9.6	0.21	0.53
	CP, g kg <sup>-1</sup> DM	157	146	4	0.03
	NDF, g kg <sup>-1</sup> DM	470	483	11	0.28

Botanical analysis of the herbage samples collected in August showed that *Agrostis capillaris* was the most important grass species, constituting between 30-50% of the fresh weight of the grass fraction, followed by *Festuca rubra* at 10-35%. It was more difficult to identify the most common forbs selected. Forty-eight species were identified. Among the most common were *Trifolium repens*, *Anthriscus sylvestris*, *Trifolium medium* and *Achillea millefolium*.

## Conclusions

No significant differences in vegetation type, energy content or NDF content were found between the herbage selected by steers grazing a semi-natural pasture that had been left ungrazed the previous season compared with the herbage selected in a pasture grazed the previous season. A slightly higher CP content was nonetheless observed in the pasture that had been left ungrazed the previous season. It can therefore be concluded that the quality of herbage available for cattle on semi-natural pastures left ungrazed the previous season is comparable to the quality of semi-natural pastures grazed yearly. However, it is important to emphasize that long-term changes in sward composition were not part of this study.

## Acknowledgements

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# Improvement of oligotrophic pastures with animal and mechanical treatments

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## Abstract

Reduction of the great sheep transhumance, which in the past was important over a large area of summer pastures in Central Italy, has caused the diffusion of oligotrophic herbaceous species and adversely affected pastoral resources. The main objective of this work was to evaluate the effects of some pasture improvements in an area dominated by *Festuca paniculata* and *Brachypodium genuense*. Decreased grazing pressure has the tendency to lead to these species dominating the sward, with reduction in floristic richness, biodiversity and forage quality. In order to improve the pasture in terms of productivity and forage quality, the effects of animal grazing (horses and sheep) and of mechanical treatment (presence or absence of herbage cut) were studied on a natural pasture. The experiment was carried out over three years and data collected on the presence of the above-mentioned species and the pastoral value of the pastures. Results showed that horse grazing was very successful in reducing the presence of *F. paniculata* and *B. genuense*. However, cutting was rather ineffective in enabling the pasture to recover and it was much more expensive.

Keywords: *Festuca paniculata*, *Brachipodium genuense*, cutting up, horses, sheep

## Introduction

Transhumance was practised in the past in many parts of central Italy and was mainly carried out using sheep from the lowlands of the South for the summer utilization of the inner mountain pastures of the Apennines. This practice is nowadays greatly reduced (Talamucci and Pardini, 1999) and the decrease of grazing of pastoral resources affects many characteristics of the pastures and their structure (Staglianò *et al.*, 2003). A very important change that can be observed as a consequence of decreased animal utilization is the development on the pasture of oligotrophic herbaceous species, which are characterized by a strong ability to grow in soil with reduced fertility. Examples of oligotrophic species in many Italian situations are *Brachipodium* spp., *Bromus erectus* and *Festuca paniculata*, which are very common under dry conditions and which are considered as high competitors in poor environments where the retention of available resources is a primary strategy (Wilson *et al.*, 1999). Establishment of these plants produces many undesirable effects in terms of botanical composition, quality of the offered forage, floristic richness and biodiversity (Messerli *et al.*, 2007). In order to evaluate how mechanical treatments or different kinds of animal can be used to recover a plant community dominated by oligotrophic species, an experiment was carried out in the period 2000-2002 in a pasture located in an area of central Italy.

## Materials and methods

The area belongs to the Chiarano-Sparvera reserve, a public area managed by the National Forest Service in Abruzzo (Central Italy). The experimental site consisted of a sector of a natural pasture on which a remarkable presence of *Festuca paniculata* and *Brachypodium genuense* had developed due to the reduced stocking rate of last decades. The total surface of the experimental site was 4 ha, 2 ha of which were fenced. The 2 ha fenced plot was grazed

by 10 horses for 15 days at the beginning of the season in the years 2001-2002 with an average stocking rate of  $0.20 \text{ LU ha}^{-1} \text{ y}^{-1}$ , while the other part was left to the normal utilization, i.e. sheep grazing with an average stocking rate of  $0.18 \text{ LU ha}^{-1} \text{ y}^{-1}$ . Inside each main plot, 8 subplots of  $2500 \text{ m}^2$  were identified, and in half of these the existing phytomass was cut in 2000 while the other half did not receive any mechanical treatment. According to this arrangement, the experimental design was a split-plot with four replications in which animal utilization was the whole-plot factor, and presence or absence of mechanical treatment the subplot factor. Data collection over time was conducted through linear analysis according to Daget and Poissonet (1971) in order to evaluate the effect of the treatments on botanical composition and on pastoral value (PV), a parameter which summarizes synthetically the forage value and the productive potentiality of a pasture (Cavallero *et al.*, 2002).

## Results and discussion

Animal grazing affected in a remarkable way the occurrence of the two main oligotrophic species existing on the pasture. There was a relatively large presence of *Festuca paniculata* in the pasture at the beginning (contribution of 20.9%) but at the end of the two years of horse grazing its frequency was greatly reduced to less than half that in the area grazed by sheep (Figure 1). Presence of *Brachypodium genuense* was also positively affected by horses, and in 2002 it had almost disappeared. The effect of cutting was significant on *Festuca paniculata*, but in this case a reduced contribution of this grass was observed in the uncut plots, and we can argue that in this situation the cutting stimulated its growth. By contrast, there was no significant effect observed for *Brachypodium genuense*.

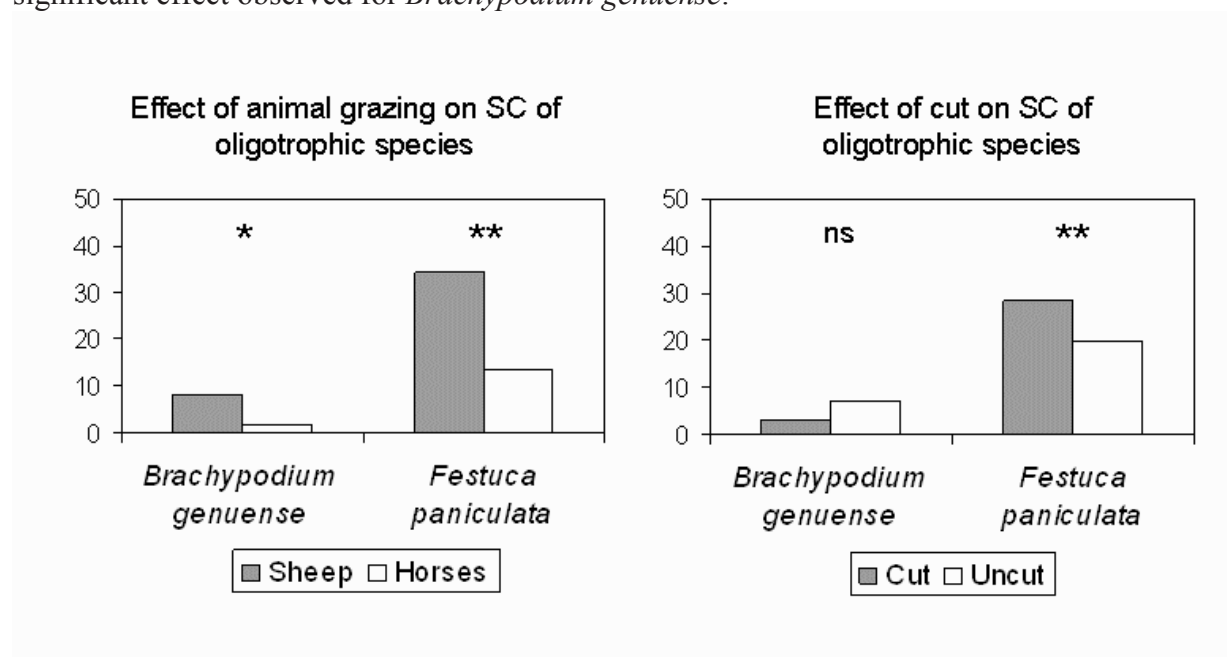


Figure 1. Effect of animal grazing and cut of phytomass on specific contribution (SC) of *Brachypodium genuense* and *Festuca paniculata* at the end of the experimental period. ns = not significant, \* = significant difference ( $P < 0.05$ ), \*\* = highly significant difference ( $P < 0.01$ ).

The effects of the treatments observed for these two species also greatly influenced the pastoral value. Differences for this parameter became increasingly evident over time, and by the second year a difference of more than 11 in PV was observed between plots grazed by horses and those grazed by sheep (Figure 2). There was no significant effect of phytomass cut in 2001 but a highly significant difference was detected in 2002, with higher values from uncut plots. Thus, the effect of treatments on pastoral value followed nearly the same trends observed for the specific contributions of the undesirable grasses.

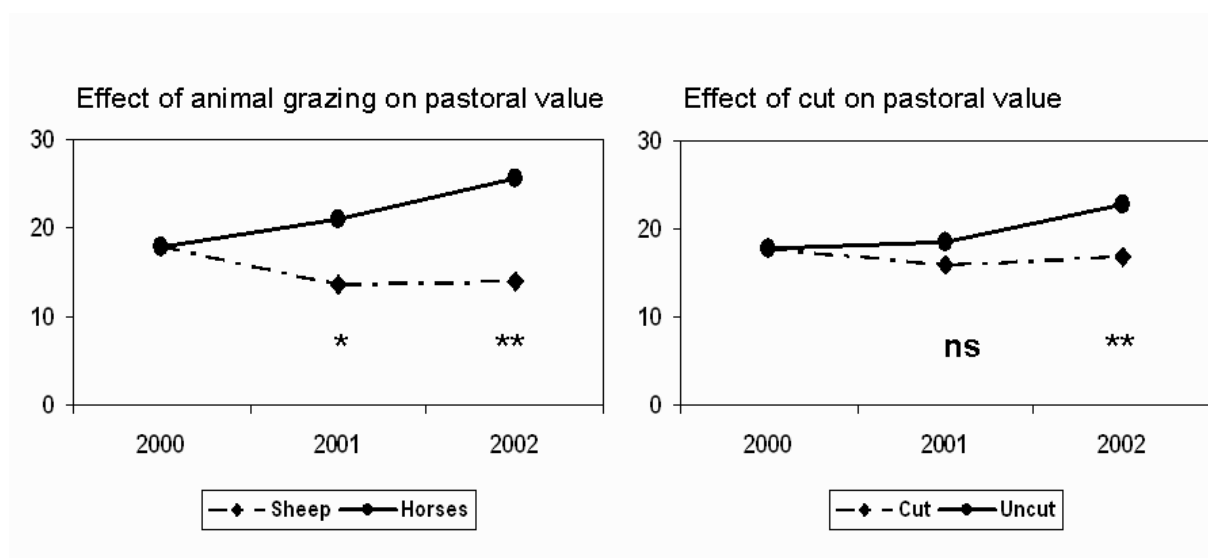


Figure 2. Effect of animal grazing and cut of phytomass on pastoral value during experimental period. ns = not significant, \* = significant difference ( $P < 0.05$ ), \*\* = highly significant difference ( $P < 0.01$ ).

The results showed the possibility for using horses to recover pastures that have deteriorated through the ingress or expansion of herbaceous species that have a reduced palatability. However, herbage cutting produced contrasting results, both in terms of occurrence of undesirable grasses and in its effect on pastoral value.

## Conclusions

The use of horses seemed to be a more certain method for the rehabilitation of the area than the mechanical treatment which, in some cases, produced ineffective results. However, the use of animals can be a key action only if conducted properly, with a very high instantaneous stocking rate for a limited period of time, in order to depress the animal selectivity and to produce effective utilization of species of reduced palatability, such as those encountered in this area. Concentrated grazing should be imposed at the beginning of the vegetative season to depress root reserves, especially in the case of *Festuca paniculata*. Further studies on this topic should investigate whether grazing with sheep could be as effective as horses. These improvement techniques should be planned only if there is the real possibility to increase the animal stocking rate afterwards, in order to maintain and preserve the improvements obtained.

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# Effect of two different supplementary feed rations on pastures in an area of the Italian Alps

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## Abstract

The present work focuses on a trial conducted on the pastures of Malga Juribello located in the Paneveggio-Pale di San Martino Regional Park. The aim of the experiment was to investigate interactions between the quantity of feedstuff supplied daily to livestock and some important pastoral features. The trial area was divided into two plots. In one area (H) the animals received a higher feedstuff supplementation, whereas in the other area (L) the livestock received a lower ration. Data collection on pastures concerned botanical analysis, production and height of *Deschampsia caespitosa*. Results showed a positive effect in the L plot in terms of presence of legumes and pastoral value. In these conditions, even less palatable species, such as *Deschampsia caespitosa* were grazed, permitting a positive evolution of the pastures. This demonstrates that an extensive management could result in a better pasture quality and that it can contribute to the preservation of delicate habitats.

Keywords: pastures, grazing, feed ration, *Deschampsia caespitosa*

## Introduction

In many summer pastures of the Alpine chain some local breeds of animals, which are not highly productive, have been substituted by more specialized breeds characterized by high feeding requirements and thus requiring large amounts of feedstuff (Cozzi *et al.*, 2004). Nevertheless, particularly in the most difficult environments of the mountains, these kinds of animals are not very suitable for grazing and for this reason the pastures are often undergrazed. This situation can lead to undesirable consequences, such as degradation of vegetation, floristic simplification, oligotrophic species encroachment and, sometimes, to eutrophication (Zimkova *et al.*, 2007). The present study reports the comparison of some pastoral characteristics as affected by different feed supplementation of animal grazing.

## Materials and methods

The trial site was represented by 'Malga Juribello', an area inside the Regional Park of Paneveggio-Pale di S. Martino (Veneto, North Italy) on 270 ha of pastures dominated mainly by *Nardus stricta* and *Deschampsia caespitosa*, and located at 1820-2230 m a.s.l. (Orlandi *et al.*, 2000). The experimental trial was carried out on an homogeneous surface of 24 ha split into two plots of 12 ha each. Each section was grazed during summer by a herd of 12 cows of Brown Italian breed that received an high (H) or a low (L) level of supplementation of feedstuff represented by 6 kg day<sup>-1</sup> for the group H and 2 kg day<sup>-1</sup> for the group L. The feeding took place twice a day at milking, which was conducted directly on the pasture with a mobile machine. The trial was carried out for three years and all the following results concern data gathered at the end of this period. Data collection concerning vegetative composition was performed for each section on 8 circular sample areas with the diameter of 4 m. In each area all the occurring species were identified and an index of abundance was applied to each species according to Pignatti (1976). Botanical data permitted the evaluation of the specific contribution of each species (SC) and of the pastoral value (PV) according to Daget and



Poissonet (1971) and of the biodiversity by the Shannon index ( $H'$ ) (Magurran, 2004). Moreover, in the same sample areas, the height of each stem of *Deschampsia caespitosa* was measured to evaluate the effect of grazing on this grass which is in a phase of vigorous spreading. Eight exclusion cages ( $1\text{ m}^2$ ) located on each section cut at the end of the growing season permitted the evaluation of the DM production, inside the cage, and the utilization rate, by the ratio between the herbage utilized by animals (difference between biomass data collected inside and outside the cages) and the DM production. The results were analysed statistically and means were compared using Student's t test.

## Results and discussion

In both sectors the vegetation was very similar and the two main species showed no significant differences (Table 1). In total 81 different species were counted without difference in the floristic richness of the two areas, while the average number for the sample was significantly higher in the area with a reduced feed supplementation. The L area was characterized also by higher values of Shannon index (2.54 vs. 2.32) and of pastoral value (24.2 vs. 20.6). From the productive point of view the compared plots performed in the same way, so DM yield and the utilization rate did not differ in a significant way.

Table 1. Main characteristics of pastoral vegetation in the two studied sections.

	L	H	Sign.
SC of <i>Nardus stricta</i>	28.7	30.3	ns
SC of <i>Deschampsia caespitosa</i>	16.8	9.8	ns
Total number of species	68	64	ns
Average number of species for sample	35.8	30.8	**
Shannon Diversity Index ( $H'$ )	2.54	2.32	*
Pastoral value	24.2	20.6	*
DM production ( $\text{t ha}^{-1}$ )	2.08	2.48	ns
Utilization rate (%)	66	50	ns

SC = Specific Contribution; ns = not significant; \* =  $P < 0.05$ ; \*\* =  $P < 0.01$ .

Occurring species have been grouped into grasses, legumes and other botanical families (Figure 1). Grasses and other species did not show any significant difference in specific contribution between treatments, whereas legumes showed a remarkable increase in the L section (8% vs. 3%). This increase was probably the main factor affecting the quality of the pasture and, consequently, the pastoral value. Particularly, the species that showed the greatest increase was *Trifolium pratense* subsp. *nivale*, whose SC varied from 2% to 6%.

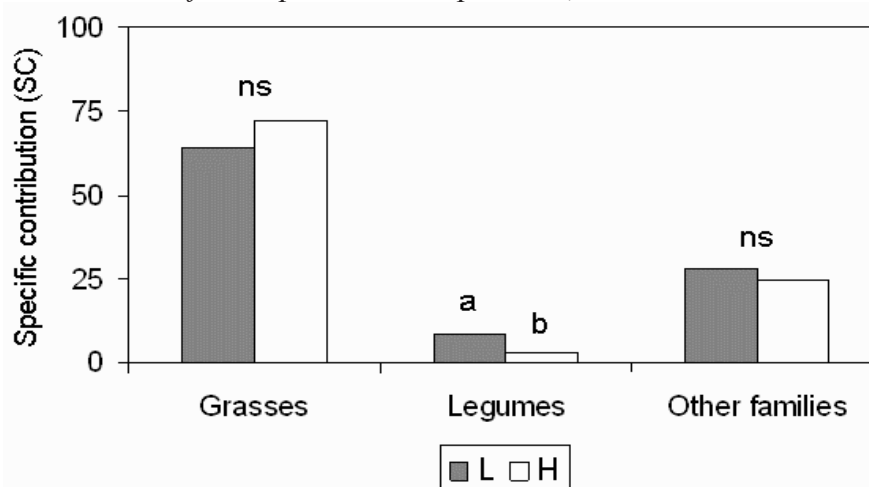


Figure 1. Botanical composition in the plots with Low (L) and High (H) supplementation. Letters (a, b) show differences between plots at  $P < 0.05$ , ns = not significant differences.



The expansion of *Deschampsia caespitosa* showed that in the H plot the stems reached a height of 17 cm, a value significantly higher than that obtained in L (12.1 cm). It seems that animals, when fed less, reduce their selective attitude towards some species, as *Deschampsia caespitosa* which is not very palatable, even if the total utilization rate of herbage is similar in the two sections.

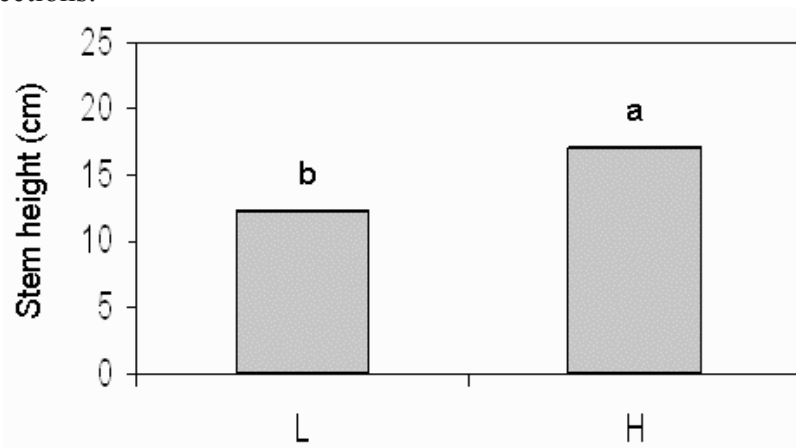


Figure 2. Stem height of *Deschampsia caespitosa* in the plots with Low (L) and High (H) supplementation. Letters (a, b) show differences between plots at  $P < 0.05$ .

## Conclusions

This study showed that some important characteristics of the pastures, the production and frequency of main species, were not affected by different supplementation, whereas others were influenced in a remarkable way. Positive effects were observed for biodiversity and presence of legumes, with consequent improvement of herbage quality. The reduction of feedstuff available for the animal permitted also the control of intrusive and undesirable species such as *Deschampsia caespitosa* and its higher intake allowed a greater penetration of light to the soil surface. These conditions probably explain the increase of the presence of heliophytic species in the pasture, like many legumes, that contributed to the improvement of the quality. Besides these considerations, and without taking into account the productive performances of the animals, the advantages of a management with reduced supplementation can be represented by economic features, in terms of reduced feedstuff purchase, and by environmental aspects, through a reduction of the risks of pasture eutrophication.

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# Linking *Senecio aquaticus* occurrence to grassland management

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## Abstract

*Senecio aquaticus* (marsh ragwort) is a poisonous species in grasslands of various countries (e.g. Great Britain, Central European states), and its further spread onto farmland must be prevented. To reveal links between management practice and the occurrence of *S. aquaticus*, a case-control study was carried out on agricultural grassland in the northern and central part of Switzerland in summer 2005. Botanical assessments were carried out on parcels (management units) with *S. aquaticus* occurrence (cases) and on neighbour parcels without *S. aquaticus* (controls). For all parcels, we analysed the soil nutrients and the details of the management such as intensity of fertilisation and defoliation frequency. There was a high risk for the occurrence of *S. aquaticus* with low nitrogen fertilization, with a change of management intensity in the preceding 15 years, high inclination, and gaps in sward. Despite its preferences for low nutrient sites, the species was also persistent at sites with moderate to high management intensity. We conclude that it is most important to prevent the establishment of the species during phases of vegetation change. A long-term control of *S. aquaticus* can best be achieved by avoiding sward damage and by promoting dense and stable swards.

Keywords: nitrogen applied, change in management intensity, openness of sward, on-farm survey, case-control study

## Introduction

*Senecio aquaticus* Hill (marsh ragwort) is a poisonous species in grasslands of Northern and Central Europe (Great Britain, the Netherlands, Germany, Austria, Switzerland) and can occur in high local abundance. In Central Europe, *S. aquaticus* inhabits mainly wet grasslands and fen meadows, and seems to have increased its dispersion in the last five years (Bosshard *et al.*, 2003). A further infestation of permanent grasslands by *S. aquaticus* must be prevented since the species contains pyrrolizidine-alkaloids that are toxic for cattle and other livestock (Elcock and Oehme, 1982; Witte *et al.*, 1992). This paper presents data from a survey conducted in the northern and central part of Switzerland assessing the influence of management practice and site conditions on the occurrence of *S. aquaticus*. We aimed at finding a combination of conditions where the risk for the occurrence of *S. aquaticus* was either remarkably high or low. These data will allow strategies for the control of the species to be developed.

## Materials and methods

The survey covered areas in the Swiss Plateau and the Prealps, the centre for *S. aquaticus* occurrence in Switzerland. Data were collected from a total of 72 parcels (farm management units) with *S. aquaticus* occurring in 35. The design of the survey followed a matched case-control study, a standard method in health studies (Agresti, 2002). At a particular location, usually two parcels were selected: one where *S. aquaticus* was present (cases), and one where it was not (controls). Both parcels had, as far as possible, similar environmental conditions; however, the management practice between them may have differed. In each parcel, we selected a representative plot of 5 x 5 m<sup>2</sup> in which all measurements were taken. We recorded

the inclination and took a representative sample of the topsoil (10 cm deep) for the analysis of soil nutrients and soil texture. A vegetation assessment established the percent cover of the species and the gaps in the sward. The farmers provided data on the amount of plant-available nitrogen applied per year, and we further registered the type of grassland management (mowing or grazing), changes in management intensity over the last 15 years (fertilizer application and defoliation frequency), and disturbance events over the last 15 years.

The influence of the recorded parameters on the occurrence of *S. aquaticus* was analysed using multiple logistic regression, the response variable being the presence or absence of *S. aquaticus*. The recorded variables were tested with forward selection and the approximate relative risk was calculated. The relative risk is the ratio of the probability of presence of the species for two levels of an environmental variable (Agresti 2002, p. 47).

## Results and discussion

Following the selection procedure, the five variables showing a significant influence on the occurrence of *S. aquaticus* were: nitrogen applied, decrease in management intensity, inclination, Ca soil content, and the openness of sward (Table 1). Doubling the application of nitrogen from 50 to 100 kg ha<sup>-1</sup> yr<sup>-1</sup> reduced the risk for *S. aquaticus* occurrence approximately three-fold (relative risk compared to the intercept = 0.38; Table 1), which means that the species occurred less frequently on intensively fertilized grasslands. Generally, high nitrogen application and mowing frequency promote species that are strong competitors for nutrients and light such as *D. glomerata* (Carlen *et al.*, 2002). The chance for *S. aquaticus* to germinate and establish under such conditions must be impaired. However, despite its preferences for low nutrient locations, *S. aquaticus* was still persistent at some study sites with moderate to high management intensity.

Table 1. Variables with significant effects on the occurrence of *S. aquaticus* (estimates of multiple logistic regression and the calculated relative risk).

Variable	Regression estimate	Relative risk (compared to the intercept)
Intercept <sup>†</sup>	-1.871	
N-applied	-0.020	0.38 <sup>‡</sup>
Decrease in management intensity	1.893	6.64
Inclination	4.963	2.70 <sup>#</sup>
Ca soil content	0.0004	1.49 <sup>¶</sup>
Openness: High (5-25%)	1.591	4.91
R <sup>2</sup>	0.48	

<sup>†</sup>The intercept represents grassland that received nitrogen of 50 kg ha<sup>-1</sup> yr<sup>-1</sup> and had no changes in management intensity. Further attributes were 0% inclination, Ca soil content of 3,000 ppm, and low openness of sward (< 5%).

<sup>‡</sup>Relative risk (compared to the intercept) for applied nitrogen of 100 kg ha<sup>-1</sup> yr<sup>-1</sup>. <sup>#</sup>Relative risk for an inclination of 20%. <sup>¶</sup>Relative risk for a Ca soil content of 4,000 ppm.

A decrease in the management intensity (fertilizing and defoliation frequency) promoted the occurrence of *S. aquaticus*, the risk being more than six-fold higher than with no management changes (Table 1). This indicates that in addition to the level of nitrogen, the process of sward development also influenced *S. aquaticus*. Community composition often changes following a decrease in management intensity. Species adapted to low soil nutrient availability will increase in their abundance and can replace the nutrient-demanding species (Koutroubas *et al.*, 2000). In our case, the decrease in management intensity must have caused a transient phase of vegetation development. Gaps in the sward are likely to arise during such

compositional changes, and *S. aquaticus* with its vast number of small, viable seeds took advantage of these conditions.

Inclination was a further significant factor: plots with an inclination of 20% had an approximately three-fold higher risk for the occurrence of *S. aquaticus* compared to plots with zero inclination. On agriculturally managed grasslands, steep sites generally reveal a higher risk to sward damage caused by cattle grazing or by mechanical harvest, especially on wet locations where *S. aquaticus* has its preferential habitats. The resulting gaps provide suitable conditions for seed germination and plant establishment (Silvertown and Smith, 1989). This interpretation is supported by a further significant factor of our analysis, the openness of sward. Parcels that had a high percent of uncovered soil (5-25%) had an approximately five-fold higher risk for the occurrence of *S. aquaticus* compared to parcels with dense swards.

Following our results one might argue that *S. aquaticus* should be controlled by increasing the management intensity. However, this will hardly be possible on many sites where *S. aquaticus* occurs, due to the limitations of topography, the soil conditions, or other factors. *S. aquaticus* might be controlled by other means. Our study revealed that the openness of sward and related factors (inclination, management changes) strongly influenced the species. Sward damage can be reduced by adjusting the grazing rates or by preventing grazing under wet soil conditions.

This on-farm survey followed the design of a case-control study. We showed that this approach can be very powerful as it provides reliable data in relatively short time (Suter *et al.*, 2007). Species occurrence can be linked to the management practice on farms and a wide range of environmental conditions existing over longer periods. Such data can hardly be obtained from experiments running for one or two years.

## Conclusions

We conclude that gaps in the sward strongly favoured the occurrence of *S. aquaticus*; thus, the maintenance of the parcels should have a high priority. For a long-term control of *S. aquaticus*, we suggest promoting dense swards, preventing sward damage as much as possible, and controlling the species when it first arrives on a parcel.

## Acknowledgments

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# Index of Authors





Aaes O., 762  
 Abdel-Galil M.E.-D.M., 203, 209, 586  
 Abdel-Gawad M.S., 586  
 Abdel-Tawab F.M., 317  
 Abdessemed A., 745  
 Abrahamsen R., 415  
 Acciaioli A., 583  
 Acosta B., 311  
 Adamovics A., 248  
 Adamson H., 977  
 Ahvenjärvi S., 780  
 Al Rifai M., 804  
 Albizu I., 613  
 Albrecht K.A., 774  
 Algerbo P., 681  
 Alibegovic-Grbic S., 916  
 Altobelli A., 998  
 Amaudruz M., 931  
 Amelung W., 601  
 Amghar F., 72  
 Amiaud B., 75  
 Amon T., 727  
 Andueza D., 501  
 Arabatzis G., 739  
 Argenti G., 87, 583, 946, 1013, 1016  
 Arkoub A., 72  
 Arnesson A., 822, 834  
 Arriaga H., 613, 736  
 Artetxe A., 574  
 Arvidsson K., 654  
 Asadi E., 441  
 Askegaard M., 200, 556  
 Audic C., 792  
 Aufrère J., 426  
 Autran P., 99  
 Baadshaug O.H., 955  
 Babic S., 326  
 Bachelard P., 69  
 Badenhauer I., 54  
 Baert J., 254  
 Bai Y., 507  
 Bakken A.K., 203, 444, 489, 592  
 Balezentiene L., 212, 777  
 Barszczewski J., 873  
 Bartolomé J., 138  
 Baumont R., 96, 375, 426, 501, 974  
 Bayat A.R., 780  
 Bazard C., 550, 980  
 Becherel F., 693  
 Bedia J., 147  
 Behrendt A., 940  
 Bele B., 159  
 Bellus Z., 529  
 Ben ahmed L., 72  
 Ben saha K., 72  
 Benedete H., 168  
 Berg Å., 27, 111  
 Bernard F., 245  
 Bernes G., 565, 819  
 Bertilsson J., 783, 867  
 Bezdrob M., 916  
 Biewer S., 568  
 Binias J., 983  
 Black A.D., 215, 492, 786  
 Blaettler T., 718  
 Bohac J.J., 78  
 Bohner A., 81  
 Boland T., 756  
 Bolzan A., 218  
 Bonifazzi B., 221  
 Borec A., 919  
 Bošnjak K., 447  
 Bretagnolle V., 54  
 Briemle G., 180, 580  
 Brocard V., 789  
 Brophy C., 39, 230  
 Buchmann N., 913  
 Buholzer S., 57  
 Burs W., 114  
 Busqué J., 147  
 Butkutė B., 471  
 Butkutė R., 224  
 Butkuvienė E., 84  
 Butnariu M., 162, 937  
 Cabaraux J.-F., 403, 577  
 Cagaš B., 272  
 Calsamiglia S., 736  
 Canals R.M., 66, 892  
 Carlier L., 236  
 Carlsson G., 227, 239, 562  
 Carrère P., 426  
 Casado M.A., 311  
 Cecato U., 501  
 Čermák B., 807  
 Cervasio F., 87  
 Chaia E., 562  
 Charrier X., 278  
 Christiansson A., 678  
 Ciepiela A.G., 571  
 Claupein W., 751

Clementel F., 1016  
 Cole L.J., 90  
 Collins R.P., 39, 230  
 Collomb M., 421  
 Combes D., 188  
 Combs D.K., 774  
 Cone J.W., 450  
 Connolly J., 39, 230  
 Coppa M., 400  
 Coquil X., 550, 980  
 Corler K., 93  
 Cousins S.A.O., 881  
 Couvreur S., 792  
 Critchley N., 977  
 Crosson P., 771  
 Cruz P., 93, 99, 631  
 Cuadros M., 132  
 Cupina B., 281, 284, 510  
 Da Ronch F., 221  
 Dahlström A.K.M., 904  
 Damour A., 75  
 Danielsson H., 432  
 Daugeliene N., 224, 233  
 Davies O.D., 977, 992  
 De Broca M., 816  
 De Francisco M., 574  
 De la Roza-Delgado B., 412, 607  
 De Lamo X., 132  
 De Miguel J.M., 311  
 De Paula Sousa D., 678  
 De Riek J., 684  
 De Smet S., 394  
 De Vliegheer A., 236  
 Dekker R., 406  
 Del Hierro O., 574, 613  
 Del Pozo A., 311  
 Delaby L., 550, 756, 759  
 Delagarde R., 759, 795  
 Despres S., 550  
 Deux N., 974  
 Di Leo V., 87  
 Disegna M., 928  
 Dods K., 474  
 Dotreppe O., 403  
 Dovier S., 998  
 Drennan M.J., 968  
 Dudilieu M., 426  
 Dufrasne I., 577  
 Dumont B., 60, 69, 96  
 Dunford B., 150  
 Duodu S., 239  
 Dupuy D'uby M., 816  
 Durgiai B., 718  
 Duru M., 93, 631  
 Duval C., 60  
 Dønnem I., 798  
 Eckersten H., 338  
 Eilertsen S., 604  
 Eknæs M., 768, 798, 810, 846, 849  
 Elgersma A., 388, 406, 450, 628, 657, 801  
 Ellen G., 406  
 El-Nahrawy M.A.-Z., 209  
 Elsaesser M., 580  
 Emanuelsson M., 907  
 Emanuelsson U., 3  
 Emile J.-C., 804  
 Epelde L., 613  
 Eriksen J., 200, 556, 559  
 Eriksson H., 453  
 Eriksson O., 129  
 Escobar Gutierrez A., 188  
 Estavillo J.M., 610  
 Evans E., 977  
 Evans J.G., 108  
 Fahmy E.M., 317  
 Falchero L., 400  
 Fallour D., 93  
 Farruggia A., 60, 69, 96, 501, 974  
 Fatyga J., 995  
 Faverdin P., 804  
 Fayolle A., 99  
 Fernandez Nuñez E., 102, 287, 616, 733  
 Ferrioune L., 72  
 Ferroni L., 218  
 Fiala J., 595  
 Fievez V., 361, 394, 684  
 Fillo M., 952  
 Finn J.A., 39  
 Fiorelli J.-L., 980  
 Flambard A., 919  
 Floris R., 242  
 Fois N., 242, 858  
 Foissy D., 550  
 Fothergill M., 39, 230  
 Frain M., 69  
 Frankow-Lindberg B.E., 456  
 Fraser M.D., 105, 108  
 Frelich J., 171, 807  
 Fricke T., 191, 568, 589  
 Frossard E., 197

- Frutos P., 147  
 Fustec J., 245  
 Gaile Z., 459  
 Gaisler J., 595  
 Garbisu C., 613  
 García-Ciudad A., 356, 634  
 Garcia-Criado B., 356, 634  
 Garel J.-P., 60, 69  
 Garmo T.H., 768, 798, 810, 846, 849  
 Garnier E., 99  
 Gaujour E., 75, 980  
 Gebbing T., 601  
 Genghini M., 87  
 Gierus M., 435, 462, 504, 507  
 Gilgen A.K., 913  
 Giustini L., 583  
 Glamocic D., 510  
 Godfroy M., 305  
 Golinska B., 194, 983  
 Golinski P., 194, 983  
 Gonda H.L., 831  
 González-Murua C., 610  
 Gonzalez-Rodriguez A., 813  
 Gouttenoire L., 980  
 Granger S., 816  
 Graß R., 742  
 Greef J.-M., 925  
 Gregoret R., 774  
 Griffiths B., 977  
 Gselman A., 598  
 Gubser C., 922  
 Gunnarsson C., 660  
 Gustafson T., 111  
 Gustavsson A.M., 432, 654  
 Gustavsson E., 907  
 Gustavsson M., 837  
 Gutkowska A., 114  
 Gutmane I., 248  
 Hadorn M., 329  
 Hakl J., 251  
 Hald A.B., 153  
 Halling M.A., 465  
 Hansen S., 592  
 Hansson P.-A., 660  
 Harkot W., 958  
 Harrison W., 90  
 Hartmann G.A., 57  
 Haugland E., 409  
 Havet A., 828  
 Havrevoll Ø., 397  
 Hedinger C., 922  
 Heiermann M., 727  
 Heikkilä T., 642, 675  
 Heinz S., 910  
 Hejduk S., 272  
 Helgadóttir Á., 39  
 Helmy A.A., 209, 586  
 Hendriks W.H., 450  
 Hermenean I., 144  
 Herrmann A., 435, 504  
 Hersleth M., 409  
 Hessle A., 117, 120  
 Hetta M., 819  
 Himstedt M., 589  
 Hobbs P., 727  
 Hoffmann J., 925  
 Holtenius K., 825  
 Homolka P., 986  
 Hornick J.-L., 403, 577  
 Hossard L., 93  
 Hovstad K.A., 63  
 Hrabě F., 595  
 Hristov K., 254  
 Hubert D., 99  
 HugueninElie O., 197  
 Huhtanen P., 765, 780  
 Huss-Danell K., 227, 239, 296, 468, 562  
 Huyghe C., 375  
 Hvelplund T., 441, 486  
 Häring D.A., 438  
 Högberg P., 227  
 Høgh-Jensen H., 559  
 Höglind M., 203, 397, 444  
 Hörndahl T., 681  
 Iacob T., 183  
 Ibraheem M.R., 317  
 Idler C., 727  
 Ilieva A., 254  
 Isselstein J., 123, 375  
 Istasse L., 403, 577  
 Jaakkola S., 642, 675  
 Jacobs dias F., 804  
 Jajic I., 510  
 Jakešová H., 595  
 Jalali A., 822  
 Jančovič J., 952  
 Jankowska J., 571  
 Jankowska-Huflejt H., 619  
 Jankowski K., 571  
 Jansson A., 989

Jansson J., 341  
 Jatkauskas J., 663, 687  
 Jauhiainen L., 675, 861  
 Jeangros B., 931  
 Jensen E.S., 559  
 Jensen S.K., 432, 565, 669  
 Jodelka J., 571  
 Johansen A., 592, 706  
 Johansson B., 825  
 Johnsson S., 852  
 Jonavičienė K., 471  
 Jouany C., 93, 99, 631  
 Jouven M., 96, 974  
 Jumpponen A., 227  
 Justes E., 631  
 Jørgensen M., 203, 409  
 Kadziulienė Z., 625  
 Kadziulis L., 625  
 Kammes K.L., 774  
 Karagić Đ., 281  
 Kašparová J., 595  
 Katić S., 281, 510  
 Katova A., 254  
 Kayser M., 123  
 Kelly F., 840  
 Kemešytė V., 257  
 Kennedy E., 706  
 Kenny D.A., 706, 843  
 Kiesel J., 925  
 Kirwan L., 39, 230  
 Klarzynska A., 126  
 Kleen J., 462  
 Klimas E., 212  
 Klimeš F., 260  
 Knežević M., 447  
 Nicky M., 645, 666  
 Kobes M., 260  
 Kohoutek A., 498, 595  
 Kolczarek R., 571  
 Komárek P., 595  
 Kornher A., 338  
 Krajic L., 251  
 Kramberger B., 598  
 Krebs G.L., 474  
 Kreuzer M., 39  
 Kriszan M., 601  
 Krizsan S.J., 819  
 Krnjaja V.S., 335  
 Krstić Đ., 284  
 Krug A., 730

Kryszak A., 126  
 Kryszak J., 126  
 Kuhn G., 910  
 Kulakouskaya T., 263  
 Kulik M.A., 266, 344  
 Kumara Mahipala M.B.P., 474  
 Kunz H.G., 580  
 Kunz P., 864  
 Kuoppala K., 391, 780  
 Kutnjak H., 447  
 Kühbauch W., 601  
 Kyriazopoulos A., 739  
 Langeveld H., 727  
 Larsen A., 293  
 Laser H., 275  
 Laurent C., 792  
 Lautrou Y., 792  
 Lazaridou M., 269, 739  
 L.-Baekström G., 341  
 Le Roy P., 804  
 Lekuona A., 610  
 Lemežienė N., 471  
 Lennartsson T., 27, 907  
 Leroy T., 60  
 Leto J., 447  
 Leuchtman A., 356  
 Lidback F., 837  
 Lidfors L., 837  
 Lind V., 409, 604  
 Lindborg R., 129  
 Lingvall P., 645, 666  
 Linke B., 727  
 Lipińska H., 958  
 Llurba R., 132, 892  
 Lochet J., 99  
 Loiseau P., 96  
 Lombardi G., 400  
 Lombnæs I., 397  
 Lopez J., 813  
 López-Díaz M.L., 135  
 López-i-Gelats F., 138  
 Lourenço M., 394  
 Lugic Z., 326, 480  
 Lunnan T., 415, 444  
 Lüscher A., 39, 174, 197, 438, 1019  
 Lättemäe P., 477  
 Lösche M., 435, 504  
 Macháč R., 272  
 Manninen M., 861  
 Marcheron H., 816

- Markovic J., 480  
 Marks E., 892  
 Marsot M., 816  
 Martin B., 361  
 Martin G., 93, 631  
 Martínez A., 607  
 Martínez-Fernández A., 412, 607  
 Martinez-Suller L., 855  
 Martinsson K., 654, 819, 867, 870  
 Marusca T., 144  
 Marzetti S., 928  
 Mathieu A., 828  
 Matić I., 447  
 Mattern T., 275  
 Mayer F., 910  
 Mc Evoy M., 756  
 Mc Cafferty P., 474  
 Mc Cracken D.I., 90  
 Mc Evoy M., 843  
 Mc Gee M., 968  
 Mc Lean B.M., 977, 992  
 Médiène S., 54, 141, 278  
 Mehlqvist M., 762  
 Meisser M., 931  
 Menéndez S., 610  
 Merino P., 610, 736  
 Merlin A., 75  
 Messeri A., 946  
 Meulenberg S., 450  
 Michaud A., 828  
 Michaud Y., 69  
 Micova P., 347, 595  
 Mignolet C., 980  
 Mihailović V., 281, 284  
 Mijangos I., 613  
 Mikić A., 281, 284  
 Mikic V., 510  
 Milano G.D., 831  
 Milić D., 284, 510  
 Misztal A., 483, 964  
 Mocanu V., 144  
 Moharrery A., 441, 486  
 Moisuc A., 162, 937  
 Moloney A.P., 361, 786  
 Monica B.,  
 Moorby J.M., 105  
 Mora M.J., 147  
 Morales-Almaraz E., 412  
 Moran J., 150  
 Moreau J.-C., 745  
 Morgan M., 992  
 Morris S.M., 105  
 Mosimann B.,  
 Mosquera-Losada M.R., 102, 135, 287, 616,  
 733, 1004  
 Murphy W., 168  
 Murtic S., 916  
 Müller C., 123, 669, 672  
 Müller J.,  
 Mäki M., 642  
 Möller J., 669  
 Mølmann J., 409  
 Nadeau E., 432, 762, 822, 834, 837, 852, 867  
 Nadin L.B., 831  
 Nadolna L., 995  
 Nagy G., 290, 934  
 Navas M.L., 99  
 Nepal S., 628  
 Nerušil P., 595  
 Nesheim L., 465, 489  
 Nesic Z.D., 335, 429  
 Neve N., 919  
 Nielsen A.L., 153  
 Nielsen P., 837  
 Nilsdotter-Linde N., 341, 432  
 Norderhaug A., 177, 415, 949  
 Nousiainen J., 765  
 Nute G.R., 361  
 Nyfeler D., 39, 197  
 Nylund R., 681  
 Nyman P., 338  
 Nysand M., 642, 648, 675  
 Nørgaard P., 762, 810, 822  
 O'Conchúir R., 150  
 O'Donovan M., 756, 840, 843  
 Odstrčilová V., 595  
 Ohlson M., 415  
 Ohlsson C., 663  
 O'Kiely P., 215, 492  
 Oksanen A., 861  
 O'Loughlin J., 840  
 Olsson K.-F., 456  
 Oreja A., 66  
 Oriol I., 937  
 Orosz S., 529  
 Ott H., 353  
 Oudshoorn F.W., 721  
 Ovalle C., 311  
 Owens V., 529  
 Paeffgen S., 403



- Pakarinen K., 495  
 Palladino R.A., 843  
 Palmborg C., 227, 296  
 Paplauskienė V., 471  
 Parente G., 998  
 Paris Le Clerc N., 54  
 Parr S., 150  
 Paszkiewicz-Jasinska A., 995  
 Pauly T., 645, 672, 678  
 Pedrol N., 607  
 Peeters A., 9  
 Pelikán J., 272  
 Pelve M.E., 1001  
 Perčulija G., 447  
 Pérez-Ramírez E., 759, 795  
 Perrot C., 693  
 Persson Waller K., 565  
 Petisco C., 634  
 Peyraud J.L., 759, 795  
 Pflimlin A., 693  
 Picard F., 501  
 Pihlgren A., 156  
 Pinto M., 574, 610, 613, 736  
 Plantureux S., 75, 305  
 Plöchl M., 727  
 Poetsch E.M., 299  
 Ponzetta M., 87  
 Popovici I., 637  
 Porqueddu C., 39  
 Portier B., 789  
 Pozdisek J., 498, 595  
 Prestløkken E., 768, 810, 846, 849  
 Prochnow A., 727  
 Prokopowicz J., 619  
 Pursiainen P., 391  
 Radovic J., 326, 480  
 Ramos M.E., 302  
 Randby Å.T., 762, 768, 798, 810, 819, 846, 849  
 Rasmussen J., 559  
 Rees E., 230  
 Repšienė R., 320, 622  
 Resch R., 299  
 Reulein J., 742  
 Richardson R.I., 361  
 Richter F., 191  
 Rigueiro-Rodriguez A., 102, 135, 287, 616, 733, 1004  
 Rinne M., 391, 495, 765, 780  
 Robertson D., 90  
 Robles A., 302  
 Rodrigues A.M., 501  
 Rodriguez-Barreira S., 1004  
 Rogge H., 940  
 Rosef L., 159  
 Rosenqvist H., 660  
 Rosická L., 595  
 Roumet C., 99  
 Roux M., 816  
 Ruget F., 305, 745  
 Ruiz de los Mozos I., 66  
 Rustas B.-O., 852  
 Ružić-Muslić D., 429  
 Rzonca J., 347, 498, 971  
 Saarem K., 397  
 Saarisalo E., 642, 675  
 Sacconi F., 87  
 Sæther N., 177  
 Saghin G., 183  
 Salama H., 435, 504  
 Salcedo G., 855  
 Salis L., 242, 308  
 Sammarone L., 1013  
 Samuil C., 183, 637  
 San Emeterio L., 66  
 Sánchez Chopa F., 831  
 Sánchez-Jardón L., 311  
 Sanderson M.A., 314  
 Santiago-Freijanes J.J., 287, 616, 733  
 Santrucek J., 251  
 Sărățeanu V., 162, 937  
 Sarbu C., 183  
 Sarunaite L., 625  
 Sarzeaud P., 693  
 Schalitz G., 940  
 Schellberg J., 601, 748  
 Scherer-Lorenzen M., 227  
 Schiborra A., 507  
 Schiess C., 165  
 Schmitt A., 168  
 Schmitt T., 792  
 Schneider M., 57  
 Schori F., 553  
 Schuepbach H., 165  
 Sebastia M.T., 39, 132, 892  
 Seidel K., 123  
 Seppoloni I., 1016  
 Seppälä A., 765  
 Seppänen M., 495  
 Sharawy W.M., 209, 317, 586  
 Sickel H., 177, 415

- Sickel M., 177  
 Simic A., 326  
 Sitzia M., 242, 858  
 Skjelvåg A.O., 955  
 Skuodienė R., 320, 622  
 Slachta M., 78, 171, 807  
 Slepetyš J., 323  
 Smit H.J., 388, 628  
 Sokolovic D., 326, 480  
 Soldado A., 412, 607  
 Sormunen-Cristian R., 861  
 Speranza M., 218, 928  
 Sprainaitis A., 350  
 Spörndly E., 120, 1001, 1007, 1010  
 Spörndly R., 645, 660, 678, 681  
 Šrámek P., 595  
 Staglianò N., 946, 1013, 1016  
 Stamirowska-Krzaczek E.A., 344  
 Stefanska J., 765  
 Stein S., 730  
 Steinshamn H., 397  
 Stenseke M., 724  
 Stettler M., 329  
 Stoltz E., 341  
 Stroia C., 631  
 Stybnarova M., 498  
 Suchý K., 260  
 Sundberg M., 660  
 Suokannas A., 642, 648, 675  
 Susan F., 221, 943  
 Suter D., 174, 438  
 Suter M., 197, 1019  
 Svendsen A., 397  
 Svennersten-Sjaunja K., 418  
 Svenning M.M., 239  
 Svensson R., 156  
 Svoboda L., 171  
 Svozilova M., 347, 498, 595, 971  
 Szűcsné-Péter J., 529  
 Søgaard K., 200, 432, 513, 556  
 Taimisto A.M., 642, 675  
 Tamm U., 477  
 Tamminga S., 406  
 Tarakanovas P., 257  
 Targetti S., 946  
 Taube F., 435, 462, 504, 507  
 Tava A., 400  
 Theau J.P., 93, 631  
 Theobald V.J., 105  
 Thomet P., 329, 864  
 Thorsen S., 203  
 Thumm U., 332, 751  
 Tiley G.E., 90  
 Tomic Z., 335, 429  
 Tonn B., 180, 751  
 Torssell B., 338  
 Trofin A., 637  
 Trommenschlager J.-M., 550, 980  
 Tuori M., 391  
 Udén P., 669, 672  
 Wachendorf M., 39, 191, 519, 568, 589, 742  
 Vale J.E., 108  
 Wallenhammar A.-C., 341  
 Wallsten J., 867, 870  
 Van Bockstaele E., 254, 684  
 Van den Pol-van Dasselaar A., 706  
 Van der Hoeven E., 388  
 Van der Horst H., 406  
 Wan H., 507  
 Van Laar H., 876  
 Van Ranst G., 394, 684  
 Van Schooten H.A., 876  
 Van Vilsteren D., 628  
 Vangen O., 177  
 Vanhatalo A., 391, 765  
 Warda M., 266, 344  
 Varekova P., 347, 971  
 Vargiu M., 308  
 Wasilewski Z., 873  
 Vasiljević S., 281, 284, 510  
 Watson C.A., 519  
 Vávrová L., 807  
 Vazquez O.P., 813  
 Vázquez-de-Aldana B.R., 356, 631  
 Vecchiettoni M., 218  
 Weisbjerg M.R., 513, 822  
 Velle L., 949  
 Vellinga T.V., 706  
 Ventura D., 132  
 Verdenal A., 188  
 Wever A.C., 657  
 Weyermann I.,  
 Vicente F., 412, 607  
 Vilčinskas E., 350  
 Villani G., 928  
 Vintu V., 183, 637  
 Virkajärvi P., 495  
 Wissman J., 27, 117

Witkowska I.M., 388, 628  
Wiwcharosky T., 934  
Volden H., 762  
Voss P., 435  
Vozár Ľ., 952  
Vranić M., 447  
Wredle E., 418  
Wróbel B., 114, 651, 873  
Vrotniakienė V., 663, 687  
Wu Z., 955  
Vuckovic S., 429  
Wulfes R., 353  
Wyłupek T., 958, 961

Wyss U., 421  
Zabalgogezcoa I., 66, 356  
Zajac M., 983  
Zanetti S., 174  
Zarzycki J., 483, 964  
Zekoniene V., 233  
Ziliotto U., 221, 943  
Zom R.L.G., 876  
Zujovic M.M., 335  
Zyszkowska M., 995  
Øpstad S.L., 949  
Östensson K., 418  
Østrem L., 203, 293

# Leaf water status of four legume species in response to water shortage

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## Abstract

In the coming years the expected increasing drought phenomena in the Mediterranean zone necessitates the study of the ecophysiological mechanisms to drought adaptation of forage species. The hydrodynamic behaviour of four legume species (*Trifolium angustifolium* L., *Medicago disciformis* DC., *Vicia cracca* L. and *Onobrychis caput-galli* (L.) Lam.) under water stress was estimated in a greenhouse experiment. Seeds of the four species were collected from natural grasslands of northern Greece and were sown in pots. Two months after seedling emergence two water treatments were applied: a) full irrigation (up to the field capacity), and b) limited irrigation (40% of the field capacity). During the vegetative period the leaf water potential and the relative water content were measured at seven-day intervals in both treatments. The results obtained suggest that *T. angustifolium* and to a lesser extent *V. cracca* were the two species mostly affected by the limited water supply. *Onobrychis caput-galli* and *M. disciformis* seemed to display an isohydric behaviour.

Keywords: legumes, water stress, leaf water potential, isohydric plants

## Introduction

The expected increase in drought phenomena for Mediterranean areas, as reported by the European Environmental Agency (2006), necessitates the study of ecophysiological mechanisms of plant adaptation to drought. This is of considerable importance especially for legumes, since they are valuable plant resources for livestock, providing high quality forage material, and they also enhance the soil nitrogen. However, relatively little is known about the hydrodynamic responses of these plants to environmental drought (Iannucci *et al.*, 2002). Given that the ecophysiological mechanisms, triggered under water shortage, differ among forage plants, indications of which legumes are sensitive to water deficit becomes significant. The aim of the present study was to assess the responses of leaf water potential and relative water content to water shortage in four legumes, occurring naturally in grasslands of northern Greece, and further to infer about their ability to grow and produce during drought periods.

## Materials and methods

Seeds from four legumes (*Trifolium angustifolium* L., *Medicago disciformis* DC., *Vicia cracca* L. and *Onobrychis caput-galli* (L.) Lam.) were collected in July 2005 from grasslands of northern Greece. The seeds were mechanically scarified prior to germination and a greenhouse experiment was conducted in December 2005. Seeds from each species were separately sown in pots of 0.6 m height and 0.5 m diameter, filled with a mixture of 50% soil, 10% perlite and 40% sand (medium mechanical composition, pH=7.6). When seedlings were of approximately 10 cm height above soil surface they were thinned to nine plants per pot. Two months after seedling emergence (April 2006) two irrigation treatments were applied: a) full irrigation (up to the field capacity), and b) limited irrigation (40% of the field capacity).

Ecophysiological measurements were conducted every seven days during the growing season (April to June) of 2006. At each measurement three fully expanded, mature leaves of each species were randomly selected at predawn (6:00) in order to measure the leaf water potential ( $\Psi$ , MPa), using a pressure chamber (ARIMAD-2, A.R.I. Kfar Charuv, Israel) and the relative water content (RWC, %), according to the formula described by Koide *et al.* (1991). Analysis of variance of the data was conducted with the use of the SPSS program (SPSS 15.0 for Windows). The Duncan test at the 0.05 level of significance was used to determine differences of the means among species within irrigation treatments. Regression techniques were further performed in order to explore the pattern of varying  $\Psi$  in respect to RWC.

## Results and discussion

The lowest values of  $\Psi$  were observed in *T. angustifolium* both under full and limited irrigation, while the highest values were those of *M. disciformis* and *O. caput-galli* (Fig. 1). Under limited irrigation *T. angustifolium* and *M. disciformis* showed a significant decline of 57% and 52% respectively in  $\Psi$  in relation to their values under full irrigation, while this decline was lower in *V. cracca* (33 %) (Table 1). The low  $\Psi$  values could be attributed to low water uptake, low hydraulic flow rates within the plants or high water loss rates (Iannucci *et al.*, 2002). On the opposite, no statistical differences were observed in the  $\Psi$  of *O. caput-galli* between the two irrigation treatments (Table 1); in fact its  $\Psi$  was higher under limited irrigation. As far as RWC is concerned, no differences were observed among the species both under full and limited irrigation (data not shown).

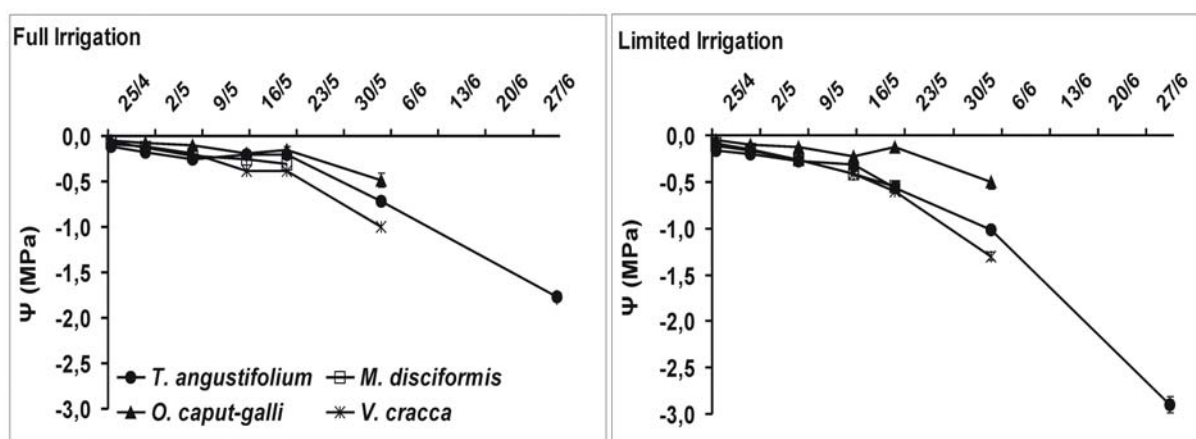


Figure 1. Seasonal change of leaf water potential ( $\Psi$ ) of the four legumes studied under full and limited irrigation.

Table 1. Leaf water potential  $\Psi$  (MPa) of the four legume species under different water regimes. The numbers represent means  $\pm$  S.E. Different letters in the same column indicate statistical differences between irrigation treatments means within species.

Irrigation	<i>T. angustifolium</i>	<i>M. disciformis</i>	<i>O. caput-galli</i>	<i>V. cracca</i>
Full	- 0.50 $\pm$ 0.13 a	- 0.20 $\pm$ 0.03 a	- 0.18 $\pm$ 0.04 a	- 0.36 $\pm$ 0.08 a
Limited	- 0.78 $\pm$ 0.20 b	- 0.30 $\pm$ 0.05 b	- 0.19 $\pm$ 0.04 a	- 0.48 $\pm$ 0.10 b

The relationship of  $\Psi$  and RWC has often been used to quantify the dehydration tolerance of tissues; tissues maintaining a high RWC with decreasing  $\Psi$  are considered more tolerant to dehydration (Iannucci *et al.*, 2002). *T. angustifolium* and *V. cracca* showed a proportional

reduction of RWC in relation to  $\Psi$  under limited irrigation (Fig. 2); in the former species the RWC decreased to almost 50% under limited irrigation, corresponding to a  $\Psi$  value of - 3.0 MPa. On the other hand, *M. disciformis* and *O. caput-galli* showed a more isohydric behaviour, since they responded more conservatively to water shortage, trying to maintain both RWC and  $\Psi$  in high levels probably through stomatal closure induced by hydraulic or chemical signals (Jones, 1998). Isohydric plants tend to adjust their stomatal behaviour in such a way as to maintain leaf water status relatively stable as environmental conditions change (Tardieu and Simonneau, 1998).

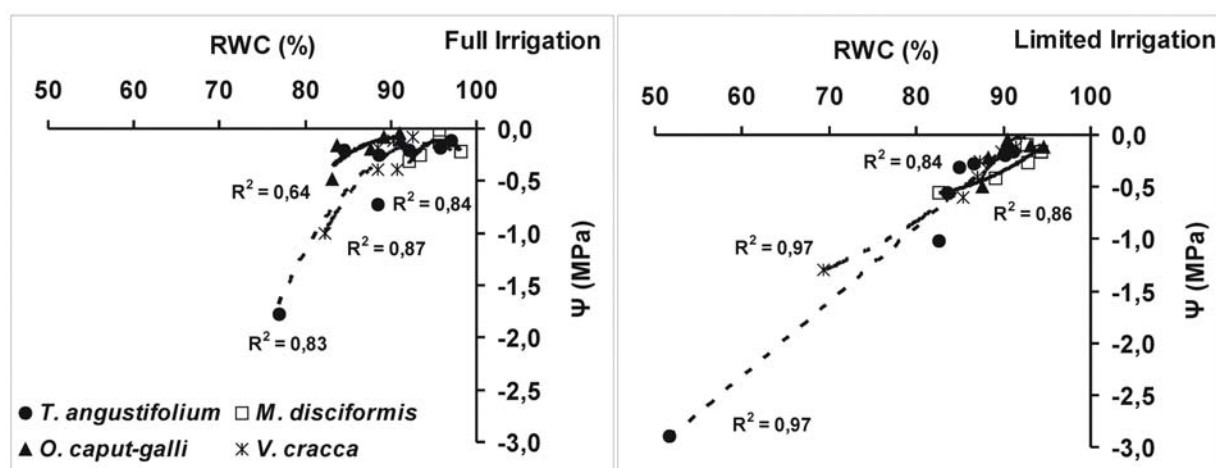


Figure 2. Relationship between leaf water potential ( $\Psi$ ) and relative water content (RWC) of four legumes under full and limited irrigation.

## Conclusions

*Trifolium angustifolium* and to a lesser extent *V. cracca* were the two species that were most affected by the limited water supply, while *O. caput-galli* and *M. disciformis* displayed an isohydric behaviour, indicating that they could be considered as less sensitive to limited irrigation.

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