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Edited by

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Proceedings of the 26th General Meeting
of the European Grassland Federation
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Foreword

The main theme of the 26th General Meeting of the European Grassland Federation is 'The Multiple Roles of Grassland in the European Bioeconomy'. But what does 'Bioeconomy' mean? According to the European Union, Bioeconomy encompasses the sustainable production of renewable resources from land, fisheries and aquaculture environments and their conversion into food, feed, fibre, bio-based products and bio-energy as well as their related public goods. Bioeconomy is an important element of Europe's answer to the challenges that lie ahead. The Bioeconomy includes primary production, such as agriculture, forestry, fisheries and aquaculture, as well as the industries that use and process biological resources. Grassland management and forage production are central elements in a bio-based economy, as is forage based livestock production.

There are five plenary sessions focusing on (1) reasons for ruminants and grassland being disconnected and strategies for reconnecting them; (2) pasture and forage potential; (3) associated quality evaluation for ruminant nutrition; (4) synergies between ecosystem services, biodiversity and agricultural production in grasslands; and (5) grasslands in a changing climate with focus on adaptation and mitigation. In the opening session grassland production in Norway will be presented in historical, current and future perspectives.

Five Mid-conference tours will be organised in the surroundings of Trondheim, presenting the diversity of agriculture in Central Norway, ranging from mountain sheep farming, lowland beef production and conventional and organic dairy farming. Different aspects of landscape, biodiversity and ecosystem services in semi-natural grasslands will also be presented. The post-conference tour will take you to the Vega Archipelago World Heritage Area, which consists of a main island and about 6,500 smaller islands and skerries. On the way to Vega we will stop at a salmon-aquarium and visit a family park in which the four largest predators in Norway (wolverine, lynx, wolf and brown bear) are to be found. Various farms and the research station Tjøtta will also be visited.

The General Meeting is organised by the Norwegian Institute of Bioeconomy Research, The Division of Food Production and Society. The division is a leading national research and competence unit with key research areas in crop production, agronomy, soil science, ecology, cultural landscape management, as well as agricultural economics and social sciences. The division has projects and assignments within plant nutrition management and fertilization strategies, agricultural and sensor technologies, environment and resource economics, impacts of land-use changes in agriculture on biodiversity and other ecosystem services, as well as projects based on systems analysis and whole value-chain perspectives.

We would like to thank all authors for their papers and presentations, numerous reviewers for their valuable remarks, members of the scientific and organising committees, the secretary of EGF and our sponsors.

We hope that the 26th General Meeting of EGF will stimulate fruitful discussions and networking, and that all participants will have some enjoyable days in Trondheim. Our aim is that the conference will be equally as successful as the General Meeting held in Ås, Norway in 1984.

Lars Nesheim	Mats Höglind	Peder Lombnæs
President	*Chair*	*Secretary*
European Grassland Federation	*Scientific committee*	*Organising committee*

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Opening session

Agricultural history of Norway 1945-2015

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Abstract

In Norway, a country with just 3% arable land and a long winter, you should not find a thriving agricultural production all over the country. To explore this agricultural puzzle, I will focus on the political and institutional development since 1945. Who were the important actors, how did they argue and how did they act? What groups were mobilised, what organisations have been active and what institutions have been built? It may be argued that the modernisation process went on more or less in the same way in all the advanced economies. However, there are huge differences in natural resources, farm structure, and political culture. Why is Norwegian agricultural history interesting for an international audience? Because the interplay between the land and the people took on quite distinctive traits here, which later became part of what might be called the Norwegian model. There are two answers to our main questions, why Norwegian farming has been thriving since 1945, in spite of its competitive disadvantages: (1) Farmers have been able to organise strong production and marketing co-operatives in order to gain more value added out of food production. (2) The Norwegian food markets have been strictly regulated, built on the regulation regime founded on a political compromise dating back to the 1930s.

Keywords: Norwegian agricultural history, farm policy, productivist agriculture, agricultural co-operatives, cultural landscape, Norwegian model

Introduction

Norway has slightly above 5 million inhabitants and 40,558 registered self-employed farmers. Norwegian agriculture consists mainly of family farms and the main farm size is 23 hectares of operated farm land. Agricultural employment constitutes around 2% of the country`s total workforce. How can it be, that in a country with little arable land, a low sun and a long winter, you find a gorgeous cultural landscape, agricultural production that is prevalent all over the country and rural people that seem to fare well? Compare this picture to some other countries in the world where you find poverty in rural areas created for wealth. There may be richer agricultural resources than in Norway, but they are less developed and less shared. Can the explanation be found at the organisational-institutional level? Can it be that the agricultural population of Norway has organised, compromised, and allied themselves in such a way that they have been able to counteract their comparative disadvantages?

To explore this agricultural puzzle, I want to focus on the political and institutional development since 1945. Who were the important actors, how did they argue and how did they act? What groups were mobilised, what organisations have been active and grew strong and what institutions have been built? What kind of alliances and compromises were formed? Farmers in Norway still speak with a powerful political voice. Why do Norwegian legislators vote for big subsidies to farming every June, when they mostly – with a big majority – favour the Agricultural Agreement between the farmers' organisations and the government?

What is the distinctive Norwegian experience from the modernisation process of agriculture after World War II? It may be argued that the process went on more or less in the same way in all the advanced economies, from the US to Europe, from France to Norway. On the other side, there are huge differences, because of differences in natural resources, farm structure, and political culture. Why is Norwegian agricultural history interesting for an international audience? Because the interplay between the land

and the people took on quite distinctive traits here, which later became part of what might be called the Norwegian model. The cultural landscapes of Norway, with little arable land but plentiful of fjords and mountains, may explain why pluriactivity has been a dominant form of production. Even with the 'death of peasantry' (Hobsbawm, 1994), part-time farming prevail, but why?

What are the turning points in Norwegian farming and farm policy after WWII? Some of them would be the same as in the agricultural histories of other industrialised countries, like the rise and fall of productivist agriculture. However, there are many traits end tendencies you will find in Norway, which will not be so prevalent in other of the advanced capitalist countries. For instance: why were the agrarian co-operatives so successful in Norway and why do they prevail? Why does the Norwegian parliament, supported by a majority in the public opinion, give so much economic support to agriculture? Compared to other European nations, the interests of farmers and rural inhabitants have an important say in decision making at the national level. Turning back to our main puzzle, how can it be, that in a country with little arable land and a long winter, there you find a beautiful cultural landscape and a flourishing agriculture all over the country? Plants have a short growing season in most of the country, and it is only small parts of the lowlands and coastal areas of South Norway that have similar possibilities for farming as our neighbouring countries. Explanations must be sought elsewhere than in soil, landscape and climate.

The 'dream of the family farm': 1945-1975

It was a slim and hungry, but not a starving nation that experienced the liberation of 8 May 1945. At the end of WWII, 3 million people lived in Norway, half of those lived in sparsely populated areas and 950,000 lived on farms. During the war years few investments were made in agricultural buildings and equipment. Nevertheless, agriculture had to wait, because priority was given to those manufacturing industries earning foreign currency through exports. In addition, the Northern Provinces were devastated because of the scorched earth tactics during German withdrawal, and needed everything from farm animals and living houses to ports and hospitals. Imported agricultural machinery was rationed. Credit was scarce, and the Government channelled financial aid through agricultural credit associations and encouraged the farmers to join tractor co-operatives.

The Labour Party now started a period of 20 years in power. The main policy goal was increased public welfare based on economic growth. To reach that goal, labour force and investment capital was channelled to the most productive industries. The primary industries; agriculture, forestry and fishing had accumulated a surplus labour force during the crisis and war times. Towns and suburbs grew quickly and rural areas were depopulated. Between 1945 and 1950 the numbers of working hands in the manufacturing industries passed the numbers of those working in agriculture. This was a symbolic event, showing that Norway had entered the era of social democratic industrialism.

A technological and social revolution in agriculture

Although agriculture was not given first priority by the Government, farmers were quite optimistic in the first post-war period. Investments were made, production increased and new markets were opened. Lack of labour force now employed by the manufacturing industries pushed farmers to invest in labour saving technologies. The first to leave agriculture were relatives and hired farm hands, and women left before men (Almås, 2002: p. 123). This exodus of agricultural labour force changed the gender system of agriculture, starting a process of masculinisation of agriculture, which has lasted up to this day. Both push and pull factors were strong. Incomes in the urban industries were higher and social benefits, like vacation, were also drawing cards. Urban housing was well organised in the first post-WWII years, offering a completely new life for rural migrants. When labour became scarcer in rural areas, demand for wage increase to urban standards trigged farmers to invest in timesaving technologies. To find seasonal helpers in labour intensive summer weeks also became more difficult. Consequently intensified mechanisation was a logical solution. In the 1950s when credit and import regulation was eased, mechanisation went on at a forced rate and pushed labour out of agriculture.

Socially, WWII was in many ways an end to 2,000 years of peasant tradition (Bull, 1979: p. 237). Alongside with the industrialisation of farming, social relations at the farm and in the countryside changed fundamentally. From being a large labour unit with several farm helper hands, most farms in a few years reduced to a household operation. Running water and electricity had revolutionised the working life of rural women. Reduced number of children and fewer working people on the farms, as well as less home processing and consumption, decreased the female labour burden drastically. The labour force on many farms was also reduced to the ownership nucleus family. This modernisation of the working role of the farmer's wife also changed her social relations. Farmwomen became more active in social life, establishing for instance a national association of farmers wives in 1946. Soon voluntary associations or municipalities started to build elderly homes to take care of people in old age. Cultural habits deeply embedded in the community crumbled. Social habits around weddings, funerals and other family events were simplified. Welfare policies were also extended to rural areas, like old age pension (1936) and children's allowance (1946).

The farm structure also started to change in the 1950s, as it became difficult to make a decent living matching the demands for income at the time. Many small farms were depopulated, or the family members started to work off the farm, just farming part-time or renting away the land. Some of those farms had been established as crofter homes of as new farms in the 1920s and 1930s. Out of 195,000 units owned by the farm family, 75% had less than 5 hectares of arable land. Only 41% of units had farming as the only source of living. Now, as the family farm was the ideal unit of farming according to Government plans, agricultural policies gave investment priority to units that had the possibility to expand. By most politicians of the time, the sustainable family farm was meant to be a permanent and central part of the rural social system. A new agricultural law in 1955 prohibited farm partition and gave the government first right to buy when cultivated land came on sale. Such land should later be sold to neighbouring farmers, as a way to strengthen their economic sustainability. This structure rationalisation, partly created by market mechanisms and partly induced by government policies, led to the liquidation of 44,000 small farms during the 1950s. The production, however, increased, made possible by tractorization and raising productivity in animal and plant production. Cheap fuel and state funded agricultural research spurred this emergence of productivist agriculture.

Within the governing Labour Party, there were cleavages and conflicts over agricultural policies. On the one side were the national economists and their allies within the Government, who wanted to transfer labour and financial capital quickly from the primary industries to the fast growing export sectors. After WWII, these people took a firm hold of the economic policies of central party and government apparatus, and remained in power up to 1965 when the Labour Government resigned. Agrarian and rural groups in the party tried to mobilise support for small scale agriculture, without success. Their power base was the Smallholders' Union and the labour group in the Parliament. As agriculture itself, this group had to fight on the defensive in the party, and lost a decisive battle in 1962 when the majority of the Parliament chose to close the Smallholders' College, following a recommendation from the labour minister of agriculture.

One way to increase the size of small farms was by the use of cheap concentrated feedstuffs and chemical fertilizer. In this way it was possible to increase the production without adding much land to the farm. From the early 1950s a policy of canalisation was introduced, which in principle paid grain better than milk and meat in relative terms. The goal was to promote a regional specialisation, in which the lowland Eastern Provinces mainly produced grains, while animal production based on grass was canalised to the fjords and valley regions and the north.

Partly because of better cultivation and use of fertilizer and chemicals and partly because of better plant material, the yields increased quickly during the first post-war decades. A network of agricultural college institutes, experiment stations, and co-operative trial rings made it possible to do applied research and spread the results to practical farming at a fast pace. This knowledge system, partly inspired by the US Land Grant Agricultural College system, became famous for the close ties between researchers and practical users. As a part of the Marshall Plan, 60 grants were giving to promising young scholars in the years 1950-1956. From 1956 and two decades onwards, the W.K. Kellogg Foundation also gave from five to six grants every year to further education within agriculture, forestry and veterinary science. Many of those receiving Kellogg grants became leading figures within their fields later, and their US experiences were published in popular booklets (Almås, 2002: p. 197).

In animal husbandry take-off came in the 1960s, as modern breeding techniques and seamen freezing technology became available. Professor of animal breeding Harald Skjervold became at a young age a leading figure in the field, as he combined the latest knowledge in genetics with computer science and well-organised animal farmers. Not least his ability to explain the breeding principles and mobilise farmers to participate in breeding programmes, gave him legendary status. In a couple of decades new synthesized races of cows and pigs were created, based on large breeding programmes and centralised research on offspring databases. The best males by heredity were chosen for seamen banks, and after insemination was applied in a mass scale to all females in the programme, it was possible from characteristics of their offspring to choose new males for breeding. The same principles from the Norwegian breeding model were later applied to establish breeding programmes for salmon in fish farming.

The cultural landscape of Norway was drastically changed in this period. From WWII to the early 1970s the rural face was given its present form. Because of mechanized cultivation, trees and bushes were cleared away and brooks and creeks were put in pipes. Because of the closure of small farms, small plots were amalgamated and old farmhouses were taken down. The landscape became regionalised because of the canalisation policy, as grain production was predominant in the Eastern Provinces and animal husbandry was more common elsewhere. Where agriculture was marginal, as in some parts of the Northern Provinces, in the mountainous areas and in the fjords, the land was not cultivated anymore and became gradually covered, mainly by deciduous forest. This process of forestation slowed down when agriculture experienced better times in the 1970s and 1980s. All through these years, the Government financially stimulated the cultivation of new agricultural land from forestland and mores, and new machinery like bulldozers and ditch diggers made it possible to increase the area of fully cultivated land. The total agricultural area decreased, however, as meadows and pastures were abandoned because of specialisation and mechanisation.

In forestry, the same modernisation was taking place as in agriculture. Because of the outbreak of the Korean War in 1951, prices on forest products increased rapidly, and stayed at a rather high level for several years. The long-time development, however, was a decrease in the price of timber, as is the case with most raw materials. The association of farmer forest owners (Norges Skogeierforbund) wanted to build their own processing industry to give them a better bargaining position against timber buyers. This motive coincided with Government industrial policies to modernise the pulp and paper industry, which was structurally scattered and financially weak. After almost two decades of fruitless efforts, the paper mill Nordenfjeldske Treforedling was opened in 1966 at Fiborgtangen in North Trøndelag. The forest owners, the state, and private capital owned the shares jointly. This was a typical 'social democratic deal' of that time. The forest owners through their association had at first a majority of the shares, which they later lost when the firm became a trans-national paper company: Norske Skog. The farmer forest owners succeeded to build their own industry, but lost control in the 1990s, when Norske Skog went into global acquisitions and the financial muscles of the forest organisations were too weak.

Post-WWII Norwegian productivist agriculture was built to serve the internal Norwegian market. Experiences from war times led to the conclusion that it was important to have national self-sufficiency of meat and milk products, and as high production of food grains as possible. The national market was mainly developed by farmers' co-operatives, which grew rapidly and extended their links to all regions. In the Agricultural Agreement, which from 1951 was made every year between the State Government and the two Farmers' Unions, the co-operatives were given an important semi-public role as price regulators and stock keepers. In the poultry industry, which grew rapidly from the early 1970s, both private and co-operative businesses took their share of the processing industry.

On several occasions in the 1960s, discussion on the entrance of Norway to the European Common Market came up. French president Charles de Gaulle, however, ended the discussion at both occasions, by using his veto to potential entrants. In 1971-1972, however, there was no veto, and real negotiations were held. When a majority of 54% voted no in the referendum of September 1972, one important reason was concerns for Norwegian farmers and their problems to compete with more efficient farmers in continental Europe. The mobilisation against membership in the Common Market was based on a 'green wave' in the early 1970s. This mental shift in favour of rural groups and their interests, also gives a context in which to understand the Parliamentary decision three years later to give farmers equal incomes with industrial workers. The dream of the sustainable family farm, which was dreamt by farmers and leading politicians in the post WWII decades, was farther away than ever. Like in other industrialized countries, the economic treadmill forced the farmers to run faster the faster they ran (Cochrane, 1958).

From state guarantied equity to market based diversity: 1975-2000

In the beginning of the 1970s, Norwegian agricultural policy came to a crossroad. The policy of structural rationalisation, supported by dominant political parties, as well as the farmers' organisations and co-operatives, came under pressure from different social groups. 'Fewer and stronger' as the chairman of the Farmers Union had formulated his vision didn't materialise (Færre, men sterkere' or 'Fewer and stronger' was formulated by Hans Haga, the chairman of the Norwegian Farmers Union, see Almås, 2002: p. 232). Instead farmers and rural interests lost ground concomitantly with structural changes in agriculture and rural population decline. The growth of manufacturing industries in rural areas came partly at the expense of agricultural labour force and farmers' arable land. Farmers' income was slightly more than half the income of industrial workers. Women were leaving agriculture for the growing rural labour market in the public service sector, mainly in health care and education. Farmwomen at the same time criticised their low income and marginal status, demanding a greater say in on farm and off farm decisions.

Productivist agriculture, now at the peak of its performance, was met by criticism from scientists and environmentalists, claiming that the present system lead to pollution of waterways and loss of biodiversity. The struggle against Common European Market membership in 1971-1972 had created a new understanding between centrist rural movements and urban intellectual radicals and environmentalists. For the first time since 1945, an opposition to the productivist regime of agriculture was formed. The short grain crisis in 1973, after the formation of the OPEC oil cartel and following oil embargo, was one of the decisive factors behind. Another catalyst was the 1974 UN food conference in Rome, which concluded that every country had a responsibility to produce its own food. Norwegian self-sufficiency was at that time 40%, and the UN decision came timely for those who wanted a more agricultural friendly policy in Norway.

Structural changes and populist political response 1976-1982

In the early 1970s, Norway still had relatively small farms with an average size of 6 hectares. Compared to our Nordic neighbours, only Finland had the same small-scale structure. The structural changes were rapid, however, peaking in the late 1960s and early 1970s, mainly because the rest of the economy was booming. Agricultural productivity was also increasing as a consequence of mechanisation, specialisation and growing use of inputs like chemicals, artificial fertilizers, and concentrated feed. Increasing yields in plant production as well as higher yield per animal as a result of the new scientific breakthroughs in agronomy and animal breeding, also contributed to the increased productivity. The new national division of agricultural labour contributed to increased productivity, with specialised grain production in the Eastern provinces and specialised animal husbandry in the uplands, fjords, and valleys. This increased animal production on small farms in marginal agricultural regions was partly based on grass production on new cultivated land and partly based on imported feedstuffs.

Milk production in combination with meat production was the main production outside the best grain producing areas in South Norway. The cattle races most common in Norway have been dual-purpose breeds, suitable for both milk and beef. This combined breeding policy was followed up in the 1950s and 1960s through the scientific breeding programme to create a new synthetic cow, 'Norwegian Red Cattle' (NRF). This new race was more suitable for concentrated feed and cultivated gracing land, while most traditional Norwegian races have been adapted to summer pastures in the woods and mountains. In the 1960s most farmers in Norway gave up many-sided husbandry. They specialised themselves, either in plant production, in combined milk and meat production, or in specialised production of eggs, pork or broilers. The latter productions were based on use of concentrated cheap feed, of which 52% was imported in 1973 (Almås, 2002: p. 257).

The modes of production were quickly transformed into larger units in the 1960s and early 1970s, which raised a farm policy discussion on the issue of regulation of the surpluses. In 1974 the Parliament passed a concession law, which stated a maximal number of how many pigs, hens and broilers each unit was supposed to have. The urge for this law came both from the Farmers' as well as from the Smallholders' Union, who were afraid that the rapid structural development threatened their very existence. They also got support from concerned environmental groups, which were afraid that the big units would pollute air and waterways.

From the late 1960s, summer livestock pasturing in the mountains (setring) was gradually given up, partly because of lack modern equipment and roads, and partly because of fear that animal yields would decrease with traditional methods. Later in the 1980s and 1990s, however, roads were built into the mountain valleys. The land of the valley bottoms was cultivated with mechanised methods and modern milking and cooling equipment was installed. A government subsidy to stop the decline of summer mountain pasturing was put in place, which for the time being has stabilised the number of summer pastures at around 1,300 units. At the same time there are 1000 farms participating in co-operative summer pastures of the modern type, which originated in the late 1960s as a response to lack of modern equipment in the old mountain barns. With hired hands to take care of the summer milking, the farmer could concentrate fully on the harvest chores.

However, from the 1970s and onward, free roaming sheep herds were consuming most of the grass production in the mountainous regions of Western and Northern Norway, as well as in the upland valleys of the Eastern provinces and Trøndelag. Previously holding sheep was usual on most farms, but in the 1970s specialised sheep meat production grew as in other productions. Artificial insemination was not successful as it has been in cow and pig breeding. Instead sheep owners joined rings cooperating to hold the best rams. Recently this sheep meat production based on summer pasture in the woods and mountains is threatened by a growing number of large carnivores (bear, lynx, wolverine and wolf), mainly in the border counties with Sweden. From the mid-1990s it has been a deliberate national policy in Norway to reintroduce these animals, because they were supposed to be endangered species.

As the pace of structural development was escalating, the number of part-time farmers increased. In 1970, only 1/3 of the farmers were fulltime farmers. However, this was not a new situation in Norway, as farmers also have been fishermen in the coastal districts and loggers and hunters in the forest and mountain areas. In addition, four out of five Norwegian farmers had their own forest land, which gave additional income opportunities in winter time. The new labour situation from the 1970s and onwards, was the increasing number of farmers and farmers' wives who took up full-time or part-time wage labour in the manufacturing industries and in the services. By doing so, most farm families started to scale down and simplify their agricultural production. Many even rented away their land, and in just a couple of decades neighbours rented one third of the cultivated land. Those who had land suitable for plant production grew grain or hay for sale. Other family members than the farmer left many family farms to enter the labour market. Left at home and working alone was the farmer, generally a man, who in this way became what I call a 'one-man farmer' (Almås, 2002: p. 253).

In 1975, there was an increasing unrest in rural areas, partly in response to the quick structural changes and partly in despair of low incomes and less public welfare arrangements compared to other social groups. A public commission (Øksnesutvalet) had evaluated the Norwegian agricultural policy, and in their recommendation before the Parliament in 1974 they proposed increased Norwegian food self-sufficiency. The main goal of the new policies was to target measures to small farms in marginal areas where there were few alternative income opportunities. One of their proposed policy measures were subsidies neutral to production, recently known as decoupling in European Union jargon on Common Agricultural Policy (CAP) reforms. Even though the proposed decoupling was not implemented in a big scale, one tenth of the subsidies were decoupled in the years to come. For the first time in the history of Norwegian agriculture, nutrition policy was mentioned and measures to the betterment of people's health were developed (Almås, 2002: p. 270). Most attention during the parliamentary debate of the fall of 1975, however, was given to the income goal.

During the summer 1975, a farmers' strike had broken out on the island of Hitra in Mid-Norway. 70 farmers lead by a milk producing smallholder formulated 8 demands, of which the most important was higher producer prices, equal tax benefits and social welfare arrangements. The farmers put their tax payment on a bank account, threatening to withhold the sum until their demands were met. For the first time in recent Norwegian history farmers' demands drew intensive media coverage, with the new television medium as the leading actor. The Hitra farmers directed their demands at the Government, which they both looked upon as their main opponent and their sponsor.

The tax strike got support in wide agrarian circles, being also an indirect criticism of the efforts of the two farm unions to improve farmers' livelihoods. When the two unions understood that the strike had wide support within their own ranks, they tried to take over the most popular demands and to co-opt the leaders. A couple of months into the strike, the regional leaders of the Framers Union proposed a time limit of four to six years for an income guaranty, which stated that the income of a farmer should be the same as the income of an industrial worker. In the late fall of 1975 the tax strike was stopped, after a promise from both farm unions that they would end their support for the structural rationalisation policy. Farmer friendly groups in the parliament, led by the influential politician Berge Furre from the Socialist Left Party, lobbied for a time limit to the income equity claim. On December 2 1975 the Norwegian Parliament made an almost unanimous decision that the income for one year of work in agriculture should be the same as that of an industrial worker within 6 years.

How should such a radical decision be explained? One obvious external reason was the international situation, which had put food self-sufficiency on the agenda. It has been obvious for the urban opinion in Norway that a nation that produced less than 50% of its consumed calories was very vulnerable. Secondly there has just been found oil in the Norwegian sector of the North Sea, which was supposed to ease the financial pressure on the state budget. Thirdly the tax strike had drawn wide support and spurred heated debates in many circles, which influenced the political agenda in the Parliament. This political mobilisation in favour of the countryside had not least touched young, idealistic generation with social conscience, who wanted their own country to be in use, instead of importing grain and beef from third world countries with hunger.

Gender issues and welfare reforms

In the following six years of farm income escalation, agricultural production increased rapidly, thanks to cultivation of new land, increased use of fertilizers and concentrated feedstuffs, and investments in more efficient equipment and buildings. Farm income also rose at a quick pace, and in 1982 the Agricultural Agreement actors agreed that the equity goal was reached. The result of the escalation policy was very visible in the rural areas, both on farms and off farms. In addition to investments in land, buildings and machinery, new living houses were built or the old houses renovated. Farmers bought tractors and cars, hired carpenters and entrepreneurs and went on holidays with their family.

One of the new reforms in the escalation package was a farm relief service, which gave animal farmers Government money over the Agricultural Agreement to hire farm helpers during sickness, weekends, and vacations. This generous welfare arrangement was at the time the most advanced farmers' social welfare scheme in the world. In many cases, however, the 'farmer' on paper was the male owner, while the spouse and female owner was not eligible for welfare and health benefits. The farm wives mobilised and lobbied to have equal rights as women working in other industries. Over a number of years, from the mid-1970s and up to 1990, women in agriculture were granted a fair share of the farm's income, as well as they gained compensation for vacation, sickness, and pregnancy leave.

All those social welfare reforms were embedded in the important parliamentary decision of 1975 to grant the girls equal inheritance rights with boy (Haugen, 1998). Originally the Norwegian Allodial Act had given the oldest boy the right to inherit the farm before older female siblings. This *óðal* principle dated back to the middle Ages, when this was a common arrangement for farm freeholders in the Norse area around the Atlantic (Myrhe and Øye, 2002: Chapter 2). Some of the agricultural organisations, also the Union of Farmers' Wives, were originally against this gender reform of the Allodial Act. This must be seen as a defensive act to protect the old way of life, more than a betrayal of their own daughters. Later, when the new Allodial law was implemented all the organisations in agriculture supported the government efforts to get more girls to take over farms. This is an interesting case of Norwegian 'state feminism', where reforms are developed from above, later being embraced by the people affected.

Although women in Norwegian agriculture made big leaps forward as far as income and social welfare rights were concerned, they had still a diminutive role in agricultural organisations. In 1986 Gro Harlem Brundtland formed her second Labour government with 8 women out of 18 ministers, later called 'the women's government'. For the first time Norway got a female minister of agriculture, Gunhild Øyangen. At the outset she pushed a gender equality agenda, which gave her both friends and enemies in the sector. But soon most of the farming community changed attitude and took advantage of government initiatives to train women in agriculture for their new and more influential roles.

Environmental turn and internationalisation and in the 1980s and 1990s

A Government Rural Commission proposed in 1984 to divert some of the agricultural subsidies to off farm investments and training. This scheme was at first met by opposition and resistance in farm circles, but later in the 1980s rural development became a catchword for all rural interest groups. The Norwegian Smallholders' Union was the first organisation to support the idea of state supported rural development.

Being by far the smallest of the Farm Unions, this organisation has been more radical in its approach to agricultural policy during the last part of the 1980s. Originally the Smallholders Union was formed (in 1913) by farmers in the Liberal Party (Venstre). Later, in the 1930s, this union moved towards the left of the political landscape, while the larger Farmers' Union formed their own Farmers' Party (1920). After WWII the Smallholders' Union became a firm supporter of the Labour Party, which ended in the early 1980s when a group of radicals and environmentalists took over the union. After WWII, the Farmers' Union gradually became more independent of the Farmers' Party, which changed name to the Centre Party in 1959.

Being the largest and most pragmatic of the two, the Farmers' Union has always tried to influence though negotiations more than following a line of class struggle. However, during the early 1980s, this pragmatic line came under pressure. Under the third Harlem Brundland government, she gradually changed her policies in neo-liberalist direction and started to adapt Norwegian policies and laws to EU regulations and policies. Two important lines of conflict were drawn, one internal and one external. The Government wanted to give up the income equity policy, stressing that farmers were self-employed tradesmen, who were themselves responsible for their income. In a parliamentary decision in the fall of 1992, the income and structure policy of the escalation years in the mid-1970s was turned around. Although the income equity was given up earlier in practical policy, this was the first time it was stated in a formal decision.

Concerning international relations, the Harlem Brundland Government wanted take part in the General Agreement on Trade and Tariffs (GATT) agreement, opening up the Norwegian food market for foreign competition. The 1993 results of the Uruguay round were better than expected for Norwegian agriculture, thanks to creative use of the Blair House agreement between the EU and USA. For the first the Norwegian protection of its own food production was based on legal bases in the World Trade Organisations (WTO). However, the tariffs were supposed to be reduced further in future WTO rounds, and Norwegian agriculture for the first time experienced real competition from abroad since the 1930s.

Gro Harlem Brundland also started negotiations with the European Union, which ended in the European Economic Space (EES) Agreement of 1992. This was by her and hare allies supposed to be one step towards the final Norwegian membership in the Union. The 1994 referendum, however, showed a majority against Norwegian membership. Both Farmers' Unions mobilised against Norwegian membership, mostly because a price harmonisation from day one would have been a great shock to Norwegian farmers and food industry. The new food scares of Europe had also entered the debate, leading many urban consumers to support the 'no' side (Almpås, 2002: p. 352). The agricultural reforms of the 1990s may be summarized by the following: marketization, adapting to new international frames and a waiver of the ambitious income target.

Agricultural reforms after 2000

At the turn of the century, Norwegian agriculture is internationalised. This internationalisation has been a long process, from the mechanisation and scientification in the 1950s, via industrialisation and environmentalisation in the 1980s, to the liberalisation with the WTO and EEA agreements in the 1990s. There is no way that this spirit may be put back into the bottle, or behind closed borders for that matter. On the other hand, Norwegian agriculture is shaped by its ecological, cultural and sosio-political environment. In a long period, farming in Norway was trade on the margins. Without the catch from fishing and hunting, without the lumber from the forests, without the system of pluriactivity, living from the arable lands would have been impossible. Grain imports, still necessary in this country after decades of increasing yields, had to be financed by foreign trade. Paradoxically, you will both find reasons for protection and for keeping an open economy in food production. There is a delicate balance in Norwegian politics between those forces, and so far no Government has sold out agricultural interests completely. A strong co-operative movement and strong farmers' unions may explain why agricultural interests are listened to.

Political regulations have played and are playing an important role in Norwegian post WWII agriculture. The institutional thickness of the agricultural sector has so far been a protective cocoon against further liberalisation. However, the outcome of the present Doha round of the WTO negotiations may be the toughest test for Norwegian agriculture so far. The formula of multifunctional agriculture, which is stressing the non-trade functions like cultural landscape, biodiversity and rural employment, may not be enough to secure Norwegian farmers a competitive capacity on the home market. Even with the diversification into internal niche markets and export strategies of special products like the Jarlsberg cheese, the home market for standard meat and milk products is essential for Norwegian farmers. Jarlsberg cheese export, however, will now be phased out, as the export subsidies will not be legal in WTO after 2020. After that time, farmers' dairy cooperative Tine is going to produce Jarlsberg abroad.

In the first decade after 2000, only small reforms were undertaken in the Norwegian farm policy model. Both the centre-right government (2001-2005) and the centre-left government (2005-2013) made only small regulation adjustments: The farm subsidies were maintained at a high level and the import restrictions with relatively high customs were kept in accordance with WTO obligations. But the annual negotiations on the agricultural agreement could not prevent farmers income fell down to two-thirds of the income of industrial workers and self-sufficiency of food dropped to below 50%. Despite the highly regulated and subsidized agriculture, the number of farms fell by 3-5% each year, and the remaining farmers were better off than before but worse off than the majority of the population.

In this situation, two most right-wing parties in Norway won the election in 2013 and formed a cabinet with a neo-liberalist minister of agriculture and food. The present Government wants to make fundamental changes in agricultural laws, lowering subsidy payments to farmers, and reduce import tariffs on agricultural goods. The present government's desire is that these changes should lead to larger farms and cheaper agricultural products. This rather radical policy shift to the neo-liberal right has attracted wide opposition in rural areas, as well as in centre-left policy groups in urban areas. In 2013 there was no agreement in the negotiations between the farmers' organizations and the Government, but in Parliament the farmers got 200 million NOK extra after intervention from two centre parties. However, these important negotiations ended with an agreement with the Norwegian Farmers Union in 2015 and 2016, while the Norwegian Smallholders Union refused to sign the agreement.

Several proposed law changes, like abolition of farm price control and deregulation of concession laws will be discussed in the Parliament in this and next year. A new farm policy white paper to the Parliament will be presented to the Parliament, and important negotiations are going on with the EU and WTO. The proposed liberalisation and deregulation of the farm policy proposed by the Government is met by resistance both in the Parliament and in the public opinion. At the rural grass roots, as well as in the Norwegian public, there still is a popular support for a small-scale, multifunctional agriculture all over the county, not just big, commercial farms in the central valleys in the south.

Conclusions

During the last 70 years both the Norwegian society and the Norwegian agriculture has gone through major changes, where the present structures within agriculture and the framework that constitutes their agricultural business and work environment are different from the past years. As we move up to get an overview, ten great changes may be highlighted. First of all, the agricultural society as the Norwegian society in general has been through a profound *individualisation*. Secondly, there has been a *scientification* and *technification*, both of Norwegian society at large and of agriculture. Thirdly, agriculture has been through a period of social, economic, and political *organisation*. After World War II most farmers at the regional and local level became members of dairy and meat co-operatives. Fourthly, Norwegian agriculture went through a *politicisation* and *regulation* from the 1950s and onwards. From the Agricultural Agreement in 1951 and environmental regulation in the 1970s and 1980s, until today's extensive food safety regulation, the role of the national state in Norwegian agriculture has increased. Through all those profound and broad changes runs a fifth process of change, which is a strong *integration* of farmers and rural societies into the society at large. During WWII, even though most of the agricultural production was commodified; a major part of the production was processed further and even consumed on the farm. Around 2000 all these specialised processes are gone from the farm and the farmer is an ordinary consumer.

In addition to those five processes mentioned above, which also were present before WWII, a *centralisation* and *urbanisation* process began in the late 1940s, which still is going on. This meant both a geographic and a social mobility, people moving from the North, from the inland and from the countryside and mainly to Oslo and some of the other cities along the coast. Strong pull factors were tempting rural youth to leave the countryside for the industries and services in cities. Secondly a steady *increase in the level of living* started, which also gave farmers and rural people a better life and well-being. In 1975 the Parliament decided that farmers and industrial workers should have equal income within six years (1976-1982). In that period the welfare state was opened to the farmers as they got government payment to hire farm helpers during sickness, vacation and leisure time. *Women in agriculture also got equal rights* with men from the 1970s and onwards. The change of the Allodial Act in 1975 was of high importance, both substantially and symbolically. Sickness and maternity leave as well as leisure benefits were granted women in agriculture. Four marked women may illustrate this feminisation of agricultural politics: Gunhild Øyangen became Minister of Agriculture in 1986, Anne Enger Lahnstein was elected chair of the Centre Party (former Farmers' Party) in 1991, Aina Bartmann (Edelmann) became leader of the Smallholders Union in 1992 and Kirsten Indgjerd Værdal was elected leader of the Farmers' Union in 1997.

In spite of progress in the struggle for gender equality, women left agriculture to a larger extent than men all through the period after WWII. This process of *masculinisation* may be explained both by factors internal and external to agriculture. Internal factors like labour saving technologies, specialisation and outsourcing of processing of farm products left fewer working opportunities for women on the farms. As mentioned above, care for children and elderly was institutionalised to a large extent, and rural women 'followed the jobs' out of the household and became wageworkers

Eventually there has been a fifth tendency of *downward socio-economic mobility* for the farmers as a social group. Just after WWII most farmers were still employers, at least in cultivation and harvesting seasons of the year. Both their lifestyle and cultural capital were comparable to the middle class in towns, even with distinct differences. Lifestyle and type of social life varied though the country, the class differences being most visible in the richer eastern provinces. Most full time farmers today have a low income and as a group they have rather low social status. Through their spouses' income, however, farmers may have an average household income.

To answer our main question, why Norwegian farming has been thriving since 1945, in spite of its competitive disadvantages, there are two factors to highlight above all. Because farmers have been able to organise strong production and marketing co-operatives, in order to gain market power at the national level, they have been able to gain more value added out of food consumption than would have been the case with a completely private food industry. Secondly, the Norwegian food markets have been strictly regulated up to the 2000s. This regulation regime was founded on a political compromise between farmer' and workers' interest in the 1930s. Without this compromise between rural and urban interest, in order to protect a national food production, the Norwegian rural landscape would have looked quite differently from what it looks today. However, because of the recent changes in national and international agricultural regimes, Norwegian agriculture may experience huge structural and social transformation in the years to come.

Note

This paper is to a large extent based on Almås (2004).

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Grassland production in Norway

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Abstract

Grassland and the associated ruminant livestock production is the backbone of Norwegian agriculture, as ruminant products contribute nearly 50% of the gross income of the agricultural sector. About 2/3 of the agricultural area is used for temporary and permanent grassland, and a large proportion (40%) of the arable land is used for producing cereals that are included in concentrate mixtures fed to ruminants. The huge variation in climatic conditions, caused by the wide range in both latitude and altitude as well as in the distance to the coast, determines the land use and choice of species and varieties used in grassland. However, the dominating ley grassland species used in in almost all parts of the country are timothy (*Phleum pratense* L.), meadow fescue (*Festuca pratensis* Huds.) and red clover (*Trifolium pratense* L.). The use of perennial ryegrass (*Lolium perenne* L.) is increasing, particularly in the southwestern parts of the country. The grassland yields and forage feed quality have remained very much the same during the last decade, while the intensity in ruminant production, e.g. milk yield per cow, has increased considerably. Factors that have contributed to grassland yield stagnation are probably the increasing proportion of rented farmland by larger farm units, the increasing price ratio between livestock products and feed concentrates and the increasing cost of producing forage relative to the price of concentrates.

Keywords: natural resources, grassland management, growing season, species mixtures, structural changes

Natural resources for agriculture

Agricultural land in Norway covers about 1.0 million ha, i.e. 3.3% of the total land area, and equals 0.19 ha per capita (SSB 2016). It is estimated that it is possible to bring into use another 1.2 million ha, of which more than 50% is forest and 30% is bog (Grønlund *et al.*, 2013). Approximately 13.8 million ha (45% of the total land area) in mountain and forests can also be used for extensive grazing by domestic ruminants (Rekdal, 2014). Most of the soils have been formed after the last glacial period, i.e. 10,000 years ago. The most important soils on agricultural land are Stagnosols (24%, poorly drained soils, periodically saturated by stagnating surface water), Cambisols (21%, loamy soils formed from glacial tills), Albeluvisols (19%, marine deposited silty-clay soils), Gleysols (6%, poorly drained soils, periodically saturated by ground water) and Arenosols (5%, deep sandy soils) (Sperstad and Nyborg, 2008). The proportions indicated are for the approximately 0.5 mill ha of soil that have been surveyed (Åge Nyborg, personal communication). Other soil types cover less than 4% of the agricultural land. However, there are great regional variations. For instance, Umbrisol is the most common soil which is in the Southwestern landscape 'Jæren' as are Phaeozems in the area around lake 'Mjøsa' in Southeastern Norway, both of which important agricultural areas.

Grassland cultivation is practised all over the country, i.e. from about 58° to 71° north and from sea level to about 800 m above sea level. Thus, the climatic conditions for plant production vary greatly. The growing season, the period of the year when daily mean temperature is above 5°C, ranges between 100 and 225 days. The sum of growing degree-days, i.e. the accumulated mean temperature above 5 °C, ranges between 700 and 1,200 °C in the main agriculture areas (Skaugen and Tveito, 2004). The annual precipitation varies from less than 300 mm in the rain shadow east of the west coast mountain range to about 4,000 mm on the west coast. A striking feature of the Nordic climate is the relatively high level of daily radiation relative to mean temperature in spring and early summer (Figure 1). The daily global radiation is almost the same at Holt (69°N) and Særheim (58°N) at the beginning of April, when grass growth commences at Særheim, whereas at Holt it takes at least another 40 days before the daily temperature exceeds 5 °C. Lower insolation due to low sun elevation in the north is partly compensated by the longer daily photoperiod there. In late summer and early autumn, the global radiation is much lower relative to the temperature than in spring, and the decline in daily global radiation after mid-summer is stronger in the North than in the South.

Plant material and environment

Ecotypes and varieties of cultivated grassland species developed at Northern latitudes are strongly adapted to the environmental conditions described above. The phenological reactions of grasses and clovers are governed by both temperature and photoperiod. Varieties developed at higher latitudes cease their elongating growth earlier in late summer when the day length reaches a threshold level, as compared to varieties developed at lower latitudes (Hay, 1990). The temperature may still be high

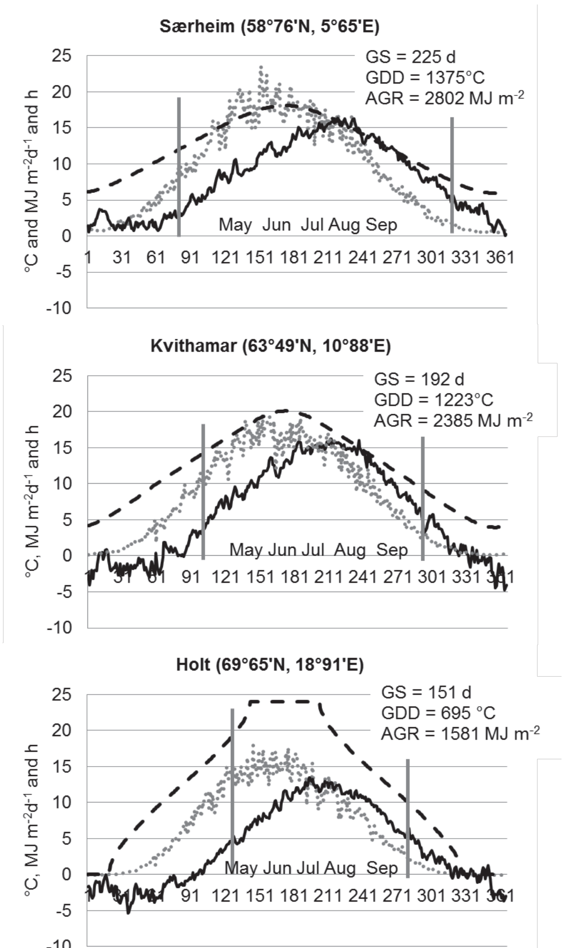

Figure 1. Daily mean temperature (°C, black line), daily global radiation (MJ m^{-2} d^{-1}, grey dotted line) and day length (h, black dashed line) at three locations during the year (Julian days). The temperature and radiation data are average across the years 1999-2011. The days between the vertical lines indicate the growing season (GS) with daily mean temperature >5 °C, GDD is growing degree days (accumulated degree sum above 5°C) and AGR is accumulated global radiation during the GS (Source: Landbruksmeterologiske tjeneste; http://lmt.bioforsk.no/).

enough for photosynthesis, but the plants invest more assimilates for respiration and survival during the winter months than for increasing growth. In spring, the relative growth rate of northern ecotypes is stimulated more by increasing day length than is that of southern ecotypes (Hay and Pedersen, 1986). Varieties developed in continental Europe are generally insensitive to the decreasing day length at higher latitudes. They continue elongation growth at the expense of accumulating assimilates and are therefore easily killed during the winter. Due to the importance of photoperiodic sensitivity, suitable varieties have been developed for most of the grassland species used in Norway. For the most important grass species, timothy (*Phleum pratense* L.) and meadow fescue (*Festuca pratensis* Huds.), Norwegian varieties are used almost exclusively. Because of their winter hardiness, varieties developed at higher latitudes are also used at higher altitudes in southern Norway.

Economic role of agriculture and land use

Agricultural production and structural changes

Agriculture accounts for less than 1% of the gross domestic production in Norway, and less than 2% of the total labour force is employed in agriculture (Rognstad and Steinset, 2012). Animal products contribute most to the value of agriculture (72% of the agricultural income). Ruminant products contribute with about 46% of the total income, with dairy milk production alone accounting for 29% and meat from ruminants for 16% of the total income (Budsjettnemnda for jordbruket 2016). The domestic production of dairy milk is relatively constant (Table 1). However, over the last decade (2003-2013), total production of beef and sheep meat (lamb and mutton) decreased by 12 and 13%, respectively, while pork production increased by 9% and poultry production nearly doubled (Table 1).

The agricultural sector has undergone strong structural changes, particularly during the last decade. For instance, from 2003 to 2013 the number of farms that keep dairy cows, sheep and dairy goats decreased by 37, 28 and 45%, respectively (Table 1). The number of these livestock groups decreased by 48, 8 and 28%, respectively. It was the number of dairy units with less than 20 cows and the number of sheep flocks with fewer than 100 ewes that decreased, while the number of larger units increased (Figure 2). These structural changes seem to be continuing.

As total milk production remained almost the same during this period (2003-2013), the annual milk yield (energy corrected) per cow increased by 21%, from 6,411 to 7,741 kg (TINE Rådgivning og Medlem, 2016). The yield increase was achieved by a 12% increase in the level of nutrition, from 33,030 to 36,853 MJ net energy lactation (NEL) per cow annually, mainly due to increased supplementation with concentrates, from 12,122 to 15,994 MJ NEL per cow annually. The silage intake increased by 17% (14,302 vs 16,694 MJ per cow and year) whereas the grazing intake decreased by 34% (5,384 vs 3,575 MJ per cow and year). Thus, the proportion of concentrates of the total intake increased from 36.7 to 43.4%.

Agricultural land use

The utilization of agricultural land reflects the importance of ruminants for agriculture's income. Temporary grassland, i.e. grassland that is kept for a few years only and then reseeded, covers about 48% of the total agricultural area, whilst permanent grassland 18% (Figure 3). Cereals and oilseed crops are cultivated on about 1/3 of the total area. Approximately 80% of the domestic cereal production is used as livestock feed, and about 50% of this is used as ingredients in concentrates fed to ruminants (Felleskjøpet, personal communication). Thus, approximately 80% of the total farmland is used for cultivating feed for ruminants.

There are marked regional differences in agricultural land use. Arable cropping of cereals and potato is mainly found in southeast and central Norway (Figure 4). The livestock density is highest in the

Table 1. Total and per capita production of livestock products per year, number of livestock farms and head of livestock heads in Norway in 2003 and 2013 (SSB, 2016).

	Total		Per capita	
	2003	2013	2003	2013
Total and per capita production				
Dairy cow milk total, ML (total) and L (per cap.)	1,482	1,448	326	287
Dairy cow milk organic, ML	17	55	–	–
Dairy goat milk, ML	20	19	–	–
Beef, Mg (total) and kg (per cap.)	85,314	83,696	18.7	16.6
Lamb, Mg (total) and kg (per cap.)	24,384	23,424	5.4	4.6
Pork, Mg (total) and kg (per cap.)	106,007	127,516	23.3	25.2
Poultry, Mg (total) and kg (per cap.)	49,043	104,030	10.8	20.6
Number of farms that keep ruminants				
Dairy cows	21,701	13,810	–	–
Dairy cows, organic				
Sheep	19,909	14,388	–	–
Dairy goats	588	321	–	–
Pigs	4,108	2,270		
Poultry	3,015	1,941		
Head of livestock				
Dairy cows total	278,137	228,895	–	–
Dairy cows, organic	5,613	8,887	–	–
Suckler cows	51,561	75,002	–	–
Sheep, >1 year old	986,639	911,211	–	–
Dairy goats	45,692	33,064	–	–
Pigs	790,717	851,058	–	–
Hens, layers	3,265,644	4,168,280	–	–
Broilers	8,108,441	15,974,607	–	–

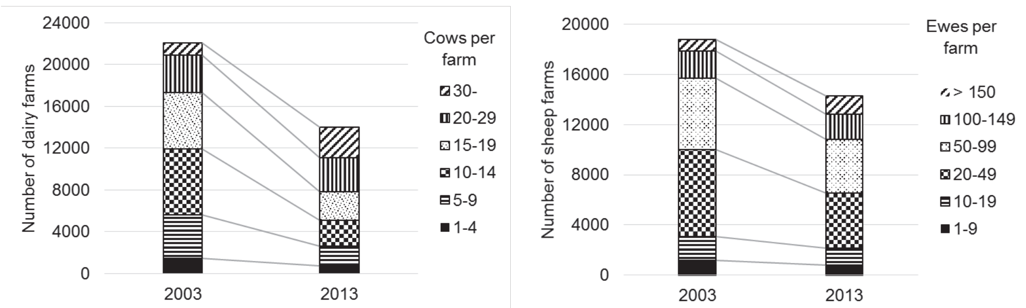

Figure 2. Changes in the total number of dairy (upper) and sheep (lower) farms and the distribution of farms according to herd and flock size (number of head), respectively, from 2003 to 2013 (SSB, 2016).

southwestern areas. This regional distribution of agricultural land use and livestock is due in part to environmental conditions, but is also largely a result of the Norwegian agricultural policy that has been employed since WWII. Important political objectives have been raising productivity and self-sufficiency,

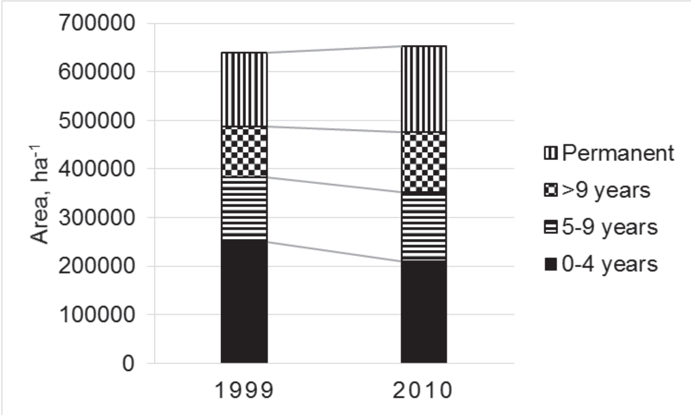

Figure 3. Grassland age distribution in Norway in 1999 and 2010 (SSB, 2016).

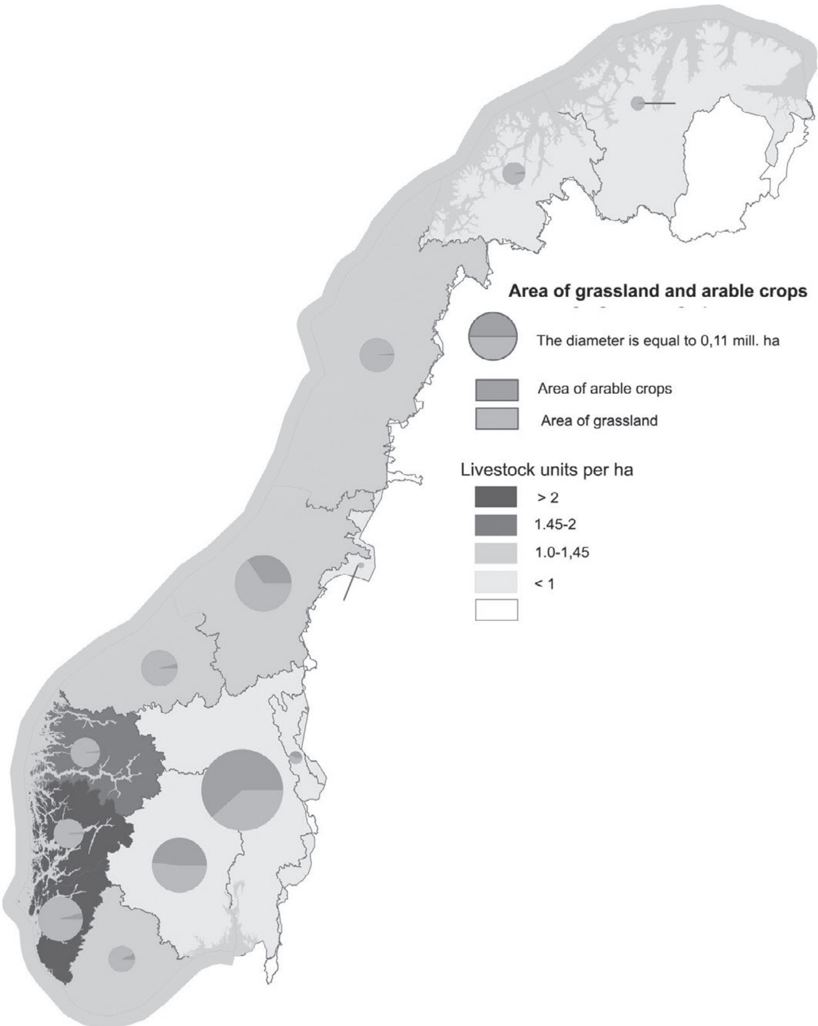

Figure 4. Regional distribution of agricultural land use as grassland and arable crops and regional livestock density. The diameter of the circle in the legend is equal to 0.11 million ha (Bye ***et al.***, 2016).

securing farmer incomes comparable to those of industrial workers, and ensuring rural employment and settlement with viable agriculture whilst maintaining and the cultural landscape (Forbord *et al.*, 2014). This has meant that arable cropping of cereals is located in areas most favourable for agriculture production, while grassland and ruminant livestock are found in more marginal areas.

Grassland management

The temporary grasslands, leys, are mainly utilized for silage production, either only cut or cut in combination with grazing. The leys are established by sowing a mixture of species, and approximately 90% of the total seed mixtures sold are used for establishing leys intended for silage production (Table 2). The most important grassland species are the grasses timothy, meadow fescue, perennial ryegrass (*Lolium perenne* L.) and smooth meadow grass (*Poa pratensis* L.) and the legumes red clover (*Trifolium pratense* L.) and white clover (*Trifolium repens* L.). Except for smooth meadow grass and white clover, all these species are introduced species. Establishment of grassland by sowing was started early in the 19th century and was not a common practice until the second half of that century (Daugstad 2008). The three species timothy, meadow fescue and red clover are used in most mixtures (Table 2). In addition, perennial ryegrass is widely used in coastal and lower altitudes of Southern Norway, often sown in pure stands. About 70% of the pure grass seed mixtures sold in 2012 (Table 2) was sold by Felleskjøpet Rogaland Agder, supplying southwestern Norway, and most of this consisted of perennial ryegrass variety mixtures. In addition, Felleskjøpet Agri sells annually around 55 Mg of perennial ryegrass seed, which is mainly used for repairing winter-damaged leys (Jon Atle Repstad, personal communication). Cocksfoot (*Dactylis glomerata* L.) and festulolium (×*Festulolium*) are used in intensive managed grasslands. In leys used for combined grazing and cutting, smooth meadow grass and white clover are included in the seed mixtures, in addition to species included in mixtures utilized only for cutting (Table 2). In dry areas, in the mountain rain shadow, *Bromus inermis* Leyss. is used, often in pure stands.

Timothy and meadow fescue account for 70 and 15% of the annual sale of grassland seeds produced in Norway, respectively (Table 3). More than 60% of the total sale of timothy seeds are of the cultivar 'Grindstad', a landrace that celebrated 100 years of use in 2015 (Nesheim 2015). 'Grindstad' is a heterogenous population that has changed over the course of time, due to changes in harvesting management imposed before seed collection.

There is a tendency that the importance of meadow fescue is declining, and that the popularity of perennial ryegrass is increasing. Norwegian ryegrass varieties are now available, the domestic seed production has increased (Table 3), and the import of seed is high. Likewise, there are domestic red and white clover

Table 2. Proportions of species in seed mixtures and total seed mixture sales (Mg) in 2012 by the suppliers Felleskjøpet Agri, Felleskjøpet Rogaland Agder and Strand Unikorn (Sources: Jon Atle Repstad, Geir Paulsen and Bjørn Molteberg, personal communication).

Seed mixture type	Species proportion in mixture, % weight basis[1]						Total, Mg
	Phleum pratense	*Festuca pratensis*	*Poa pratensis*	*Lolium perenne*	*Trifolium pratense*	*Trifolium repens*	
Silage for cutting	60-90	10-20	–	0-10	10-15	–	838
Combined silage and grazing	40-60	10-20	10-20	0-10	0-10	5-15	462
Grazing	30-40	10-25	15-30	0-10	–	5-15	59
Hay	80-90	–	–	–	10-20	–	49
Only grasses	0-100	0-30	0-20	0-100	–	–	233

[1] These figures vary both within seed mixture type (regional differences) and between suppliers and they are just indicative. For instance, Felleskjøpet Rogaland Agder uses more perennial ryegrass in their mixtures and they sell much more pure grass mixtures than the other suppliers.

Table 3. Annual sale of grassland seeds (Mg year^{-1}) produced in Norway in 2003 and 2013 (Hans Jacob Lund, Graminor AS, personal communication).

Plant species	**2003**	**2013**
Phleum pratense L.	882	1,039
Festuca pratenis Huds.	356	218
Dactylis glomerata L.	15	15
Lolium perenne L.	0	35
Bromus inermis Leyss.	30	11
Poa pratensis L.	4	34
Festuca rubra L.[1]	18	62
Agrostis capillaris L.[1]	15	12
Other grasses	12	3
Trifolium pratense L.	131	48
Trifolium repens L.	4	5
Other legumes	9	3

[1] Mainly used in seed mixtures for lawns and golf courses.

varieties, but the domestic production of clover seeds is less than the consumption so that seeds needs to be imported, mostly from Sweden and Denmark.

The age distribution of temporary grassland has changed over the last decade, during which the area of short-term leys has decreased whilst the area of leys that are renovated every fifth year or at longer intervals has increased (Figure 3).

On average, grassland used for cutting receives in total 177 kg N, 20 kg P and 107 kg P per ha annually as mineral fertilizer and animal manure (Bye *et al.*, 2016). The proportion of the total input attributable to animal manure is 0.38, 0.58 and 0.63 for N, P, and K, respectively. The application rate of both mineral fertilizer and animal manure differs greatly, and depends mainly on the length of the growing season, the number of cuts per year and the livestock density. For instance, the average livestock density is 3.3, 1.4 and 0.9 livestock unit ha^{-1} arable land in the counties of Rogaland (southwestern Norway), Nord-Trøndelag (Central Norway) and Troms (northern Norway), respectively. These are counties where ruminant production is important.

Much of the manure (57% of total) applied to grassland is spread using slurry tanks equipped with splash plates (Gundersen and Heldal, 2015). Ammonia emissions are significantly reduced when manure is applied by band spreader or direct ground injection, and the proportion of manure spread by using such equipment increased from 7% in 2000 to 19% in 2013. Most of the dairy cow manure is stored as slurry in cellars under the byres (75%) or in outdoor manure pits (23%). A larger proportion of the manure from beef cows is stored as solid dung, with 12% stored outdoors directly on the ground, 12% indoors as deep litter and 4% outdoors as deep litter.

The harvesting frequency ranges from one cut per season in marginal areas at higher altitudes and latitudes, to three and four cuts of the most intensively manged short-term leys in southwestern Norway. However, the predominant cutting system consists of either two cuts or two cuts plus one grazing per season. Nearly 80% of the harvested grass is preserved in round bales, 18% in tower or bunker silos and 2% as hay (SSB, 2016).

Of all the silage samples that were analysed for feed quality over the last three years (2013-2015), 35% were ensiled without any additives, 50% contained a formic acid based additive, 8% with combined lactic bacteria and enzyme based additive and 6% with salt based additives (Ingunn Schei, TINE Rådivning and Medlem, personal communication). The silage quality analyses show that the first and second cuts are on average fairly similar in feed value, and they are characterized by having medium levels of organic matter digestibility, crude protein and NEL (Table 4). The analyses indicate that the first cut is taken between the growth stages 'inflorescence emergence' and 'inflorescence emerged' of the dominant grass species timothy.

The statistics on grassland yields are based on estimates with a high degree of uncertainty, but they nevertheless give an indication and are presented for three counties, from south to north, in Figure 5. The figures indicate that the yields have not improved during the last decade, rather they have declined for the country as a whole. The estimated grassland yield potential is about twice as high in central and southwestern Norway and 3.5 times higher in northern Norway than the figures presented in Figure 5, even when 20% losses during harvesting and ensiling are accounted for (Bakken *et al.*, 2014). The reasons for the yield stagnation are not unequivocal. Lunnan (2012) suggested that more efficient farm operations with heavier tractors, harvesting machinery and manure spreading equipment have impaired drainage and soil structure and, consequently, grassland productivity. The farm structural changes with

Table 4. Silage quality in Norway, averaged over 2013-2015 (Ingunn Schei, TINE Rådgivning og Medlem, personal communication).[1,2]

Variable	1st cut			2nd cut			3rd cut		
	n	Mean	SD	n	Mean	SD	n	Mean	SD
DM, g kg^{-1}	14,265	317	114	8,745	333	125	926	341	118
CP, g kg^{-1} DM	14,189	152	25	8,693	161	27	915	175	27
NDF, g kg^{-1} DM	14,186	536	48	8,691	508	47	916	458	45
ODM, %	14,270	70.7	7.6	8,746	71.0	4.3	926	74.6	4.0
NEL_{20}, MJ kg^{-1} DM	14,270	5.99	0.45	8,746	6.03	0.41	926	6.33	0.39

[1] Of all the samples analysed, 76% were from round bales, 10% from tower silos and 8% from bunker silos.

[2] DM = dry matter; CP = crude protein; NDF = neutral detergent fibre; ODM = organic matter digestibility; NEL_{20} = net energy lactation estimated at standardized total DM intake of 20 kg day^{-1}; SD = standard deviation.

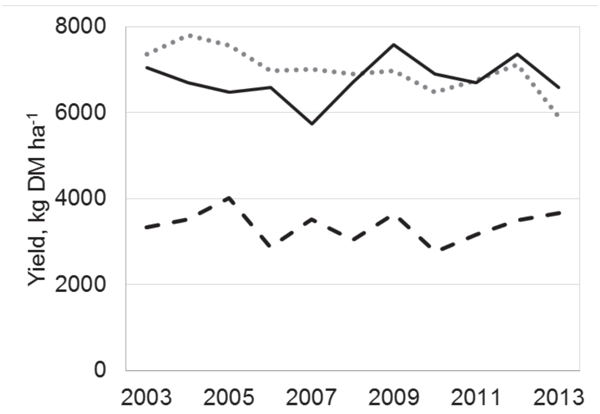

Figure 5. Average grassland dry matter yields (kg ha^{-1} year^{-1}) in the counties of Rogaland (southwestern Norway, grey dotted line), Nord-Trøndelag (central Norway, black line) and Troms (northern Norway, black dashed line) (SSB, 2016).

increased size of farm management units, where a high proportion of the agricultural area is rented, exacerbates reduced soil quality as investment incentives are low in e.g. drainage. All together 44% of all agricultural land is rented and 62% of all farm units rent land. The agriculture subsidy policy of area payments rather than payments connected to production, stimulates farmers to simply keep land in production rather than invest in management factors that improve yield. For instance, the leys are renovated less frequently than before (Figure 3). Another important reason is the cheap concentrate prices relative to product prices. For instance, the milk to concentrate price ratio increased from 1.3 in 2003 to 1.4 in 2013 (Budsjettnemnda for jordbruket, 2016). Besides, many farmers find that they have higher costs per unit NEL for their home-grown forage than for purchased concentrate (Thuen *et al.*, 2015).

Annual forage crops

Annual forage crops are cultivated on about 10,500 ha annually (SSB, 2016). On about half of this area, Italian ryegrass (*Lolium multiflorum* Lam. ssp. *italicum*) and Westerwolds ryegrass (*Lolium multiflorum* Lam. *var. westervoldicum*) are cultivated, and on the other half cereal crops harvested whole for silage, often mixed with pulses such as peas (*Pisum sativum* L), are cultivated. Italian ryegrass is mostly used for grazing, while the whole crop cereal is generally ensiled in round bales.

Woodland and mountain grazing

Traditionally, large quantities of feed were collected from rangelands, i.e. woodland and mountain areas, by grazing livestock during the summer. In terms of feed intake and production, cattle were the most important livestock used for free ranging until WWII. The importance of free range grazing has declined considerably. It is estimated that the grazing pressure of cattle, calculated as metabolic body weight ($BW^{0.75}$) per km^2, decreased from 61.2 kg in 1949 to 9.4 kg in 1999 (Austrheim *et al.,* 2011). The grazing pressure of sheep remained relatively stable during the same period and was 38.7 and 36.8 kg km^{-2} in 1949 and 1999, respectively. A total of 2.3 million livestock head were reported as grazing free range in 2014 and of these 1.9 million were sheep and lambs (Bye *et al.*, 2016).

Organic production

Currently, 4.5% of the total agricultural area is managed according to organic standards, whereof approximately 80% is used for grassland and 14% for cereals (DEBIO, 2016). As for agriculture sector in general, dairy farming is also the backbone in the organic sector in Norway, and the total organic milk production increased from 17 ML in 2003 to 55 ML per year in 2013 (Table 1). This is still not more than 3.8% of the total dairy milk production, which is lower than countries like Sweden (13%; Jordbruksverket, 2016) and Denmark (9%; Danish Agriculture and Food Council, 2016) but higher than in Finland (2%; Pro Luomu, 2016).

Challenges

The forage production costs in Norway have increased considerably during the last decade (Thuen *et al.,* 2015). Today the costs per NEL are similar to, or higher, than the market price of concentrates on many farms. The fixed costs, maintenance, machine and transport costs and depreciation, account for more than 60% of the total costs. Fertilizer is the major variable cost, with seed and silage preservation additives coming next.

High investment costs in barns associated with increased herd size and the high forage costs relative to the price of concentrate are important drivers for the increase in milk yield per cow during the last decade. A similar trend has also been seen in sheep farming. The consequence of an increased proportion of concentrates in the diet, higher dietary inclusion of imported feedstuffs such as soya and less use of pasture, is that the legitimacy of Norwegian agriculture is at stake. Continued increase in dairy milk yield per cow will reduce the grassland area needed to fill the quota, as well as increasing the import of protein feed, reducing beef production from dairy enterprises and increasing beef import, and thus it will reduce food self-sufficiency (Aas *et al.,* 2014). Intensification of animal production also raises concerns for animal welfare as well as the delivery of ecosystem services related to grazing, especially on semi-natural pasture and rangelands. Although the rather extensive, but still labour intensive, grassland farming and grazing animals are crucial for the tourist industry in mountain areas and on the western coast and in fjord landscapes, there is no common agreement as to who should pay for such benefits. Furthermore, especially on the western coast, the offshore oil and marine industries offer payments and wages that make farming, even with high subsidies and support, barely interesting from an economic point of view.

The structural changes, with increasing herd size and an increasing proportion of rented land, imposes other challenges as well. Large parts of the ruminant livestock and grassland production are located in areas where field sizes are relatively small and the distance between fields is often long. The transport distances of harvested forage from the fields to the barn and the return of manure to the fields impose considerable costs (Bergslid and Solemdal, 2014). There are examples of farmers that need to drive distances corresponding to Trondheim-Rome each year with their tractor and trailer if their slurry is to be evenly distributed on the area they manage. In practice, the fields closest to the farm receive high loads of manure whereas areas further away receive little or nothing, a situation that is of environmental concern. There are also other environmental concerns related to manure management and utilization. The springs are cold and the growing season short, and the nitrogen mineralization from manure and soil organic matter is far from perfectly synchronized with the demand of the grass crop. The window for application under optimal weather conditions is also very short in many regions. Furthermore, the small-scaled structure and long distances between farms do not allow for cost-efficient investments in biogas plants, which is often an option for better utilization of manure in other European countries.

There is of course also a discussion as to how changes in climatic conditions may challenge grassland yields and yield stability in Norway. Technologies, farm structure, ruminant diets and production systems change at a higher rate than do changes in climate and the natural and managed evolution of plant resources. It is consequently a very challenging exercise to outline what one should prepare for and how to do it. Winter-kill of grass and clover crops has always been a challenge for Norwegian grassland farmers, and there is little reason to believe that shorter and warmer, but nonetheless dark and variable, winters will in future not impose severe stress to perennial crops. In line with this, several research projects and programmes have been conducted during the last fifteen years to investigate forage crop reactions (and their genetic basis) to changed autumn and winter temperatures and precipitation patterns. For an example of this work, see for instance the recent doctoral dissertation of Dalmannsdóttir (2015).

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Theme 1.
Reconnecting ruminants to grasslands

The role of grassland based production system in the protein security

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Abstract

Ruminant production systems are under pressure for their inefficient use of land, a recurring idea assuming they consume far more plant proteins than they produce in the form of animal protein and for their methane emission. However feed-food competition does in fact concern those proteins of plant origin that are consumable by human but are actually consumed by animals and this affirmation ignores that ruminants are able to produce products of high nutritive value from grassland and marginal area that cannot be used for crop production. In addition grassland provides many environmental services. In the last decades the intensification of ruminant production system, and particularly those of dairy systems, have increased protein production per ha of land use in Europe but this occurs thanks to an increased use of concentrate which contains edible protein and at the expense of grassland acreage. Therefore the interest of this evolution for the contribution of ruminants to food security may be questioned. Feeding animals mainly from non-edible resources can be seen as a conceptually interesting issue from a global food security point of view. After showing that the decrease in European grassland acreage was closely linked to the intensification of dairy systems and the associated reduction of the number of cows, the paper describes protein production for various ruminant production systems and demonstrates that grassland based ruminant production systems are most often much more efficient than concentrate based systems for procuring proteins. The development of grassland based systems seems very desirable to increase ruminant production systems to protein security. In addition an increased use of grassland for ruminant production could also bring positive responses to societal demand for more natural practices and could contribute to maintaining farmer incomes in a context of price volatility and the provision of various ecosystem services. The challenge might be to increase productivity of grassland based system using sustainable intensification of forage production, well suited animals and grazing management.

Keywords: ,ruminants, protein production, grassland area, food security

Introduction

Livestock production provides one third of the protein consumed by humans across the planet (Herrero *et al.*, 2009) and much more in developed countries. It uses 75% of agricultural land (Foley *et al.*, 2011) of which one third is arable land and two thirds are grassland and rangeland (Steinfeld *et al.*, 2006), consumes 35% of grain products (Alexandratos and Bruinsma, 2012) and emits 14.5% of anthropogenic greenhouse gas (Gerber *et al.*, 2013). Therefore, a recurring idea assumes that livestock, particularly ruminant, is inefficient and consumes far more plant proteins than they produce in the form of animal protein and therefore livestock production is considered as exerting a significant pressure on natural resources. Since the Livestock Long Shadow report (FAO, 2006, Steinfeld *et al.*, 2006), ruminant production systems are also considered to be responsible for the emission of large quantities of greenhouse gases. Consequently, livestock production is under pressure and given this situation, many authors propose to reduce consumption of animal protein (Garnet, 2013; Eisler *et al*, 2014), mainly in developed countries where consumption is high (Bonhommeau *et al*, 2013.) for both reducing environmental degradation and improving the human population health.

From another point of view, grasslands used by ruminants are characterized by multiple functions and values which were the subject of several syntheses (MEA 2005, Peeters, 2008). They are providing forage for grazing and browsing animals, both domestic and wild. Compared with high-density coniferous tree, 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. For all these reasons, grasslands are not a crop like another. Their social and environmental importance is much higher than other crops, including other forage crops. This importance is increasingly recognized by the society and notably by the European Union (EU).

However, ruminant production systems dramatically evolved in Europe since 30 years and the use of grassland has been significantly reduced in favour to the production of silage maize and other annual crops and the use of high concentrate diet. High milk price has encouraged high-input dairy systems. The number of cows kept indoors for all or part of the herbage growing season has increased considerably in many European countries (Van den Pol-Van Dasselaar *et al.*, 2008). For forty years, dairy cow breeding has been almost exclusively oriented towards genetic potential for milk production. Today, high genetic merit Holstein cows are able to produce more than 10,000 kg milk per lactation in high-input farming systems but cannot produce such amount of milk from grazing alone. Dry matter intake and milk yield of grazing dairy cows are limited compared to conserved forage-based diets (Kolver and Muller, 1998). Similar trends were also observed for beef and sheep meat production although less marked than for dairy production. These evolutions have increased the production per unit area devoted to livestock production in Europe but in the same time European livestock became a net importer of protein as soybean meals and grains (Galloway *et al*, 2008) in spite of exporting livestock products.

Therefore several questions can be raised: Is the claim that ruminant production systems are inefficient applicable everywhere and for all type of production systems? Did the evolution of European ruminant farming systems reduce or on the contrary increase the competition between feed for ruminant and food for human? Are intensive systems more efficient to produce proteins than grassland-based production systems which also provide ecosystem services? Are grassland-based systems less profitable for farmers than intensive production systems? These are questions on which this text attempts to give some responses.

Importance of grassland areas and grassland-based systems in Europe

Surprisingly, very little studies focused specifically on the long-term evolution of the grassland area and grassland-based systems in Europe. Cropper and Del Pozo-Ramos (2006) described the evolution of livestock numbers in the EU but other information is fragmentary and rare. It was thus important to fill this gap in the knowledge of this important topic. The European project Multisward produced a first synthesis (Peeters, 2010; Peyraud *et al.*, 2012; Huyghe *et al.*, 2014) that clarifies and quantifies more precisely the importance and the evolutions of grasslands and grassland-based systems in Europe using Eurostat (2009, 2010a and b) and FAOSTAT databases. In the Eurostat database (public website), data are not available before 1990. The FAOSTAT database provides data since 1961 including for former communist countries.

Surface of grasslands and other forage crops in Europe

In the EU-27 in 2008 the total utilised agricultural area (UAA) covered 172 million hectares or 41% of the total territory. Permanent grassland covered 57 million hectares (Eurostat) (>65 million ha according to FAOSTAT) *i.e.* about 33% of UAA whereas arable land, including temporary grassland, represented

104 million hectares (i.e. 61% of UAA) and permanent crops only 12 million hectares (6% of UAA). Other surfaces including wooded areas cover 43 million ha.

The importance of permanent grasslands varies a lot between countries. Over two thirds of the of the UAA is covered by permanent grassland in Ireland (76%), the United Kingdom (63%), over half in Slovenia (59%), Austria (54%), Luxemburg (52%) and Portugal (51%) (2007). In central and Eastern Europe, the proportion in the UAA is usually lower than the European average, such as in Bulgaria (9%), Hungary (12%) and Poland (21%) (Figure 1). Romania is an exception, this country has an important permanent grassland area and its proportion in the UAA corresponds to the EU-27 average (33%). This variability reflects the differences of ecological conditions, production systems, living standards, history and policies between countries. In terms of number of hectares the United Kingdom (11 million ha), France (9.8 million ha), Germany (4.8 million ha), Italy (4.5 million ha) and Romania (4.5 million ha) are the top 5 and represent about 64% of the total permanent grassland area in EU-27.

The permanent grassland area includes about 16.9 million ha of rangelands ('poor permanent grasslands' made of grazed semi-natural vegetation) in the EU-27 territory (10% UAA in 2007), mainly in hill, mountain and Mediterranean areas. These rangelands have usually a high nature and landscape value. Spain (33.3%), the United Kingdom (24.8%), France (8.1%), Portugal (7.5%) and Italy (5.5%) include about 79% of the total rangeland area of the EU-27.

Temporary grasslands (pure grass, grass/legume mixtures or pure legume) are mixed with forage maize, fodder beat, other annual forage crops and forage legumes in the 'forage crops' statistical category. The importance of temporary grasslands varies a lot across countries. They represent 6% of the UAA in the EU-27 and 15% of the total grassland area in the EU-27. They are more important in the North of Europe (Sweden 35%, Finland 28%, Estonia and Norway 24%, Latvia 21%, Denmark 10% UAA), in Ireland (16%), in Switzerland (11%), in the Netherlands and France (10%) (Figure 1). Temporary grassland is only more important than permanent grassland in Northern Europe (Finland, Sweden and Estonia). They can also be important in some regions like in the Po valley (Italy), in Brittany (France), in the lowlands of the United Kingdom and in the Belgian Ardennes. At the opposite, in the former communist

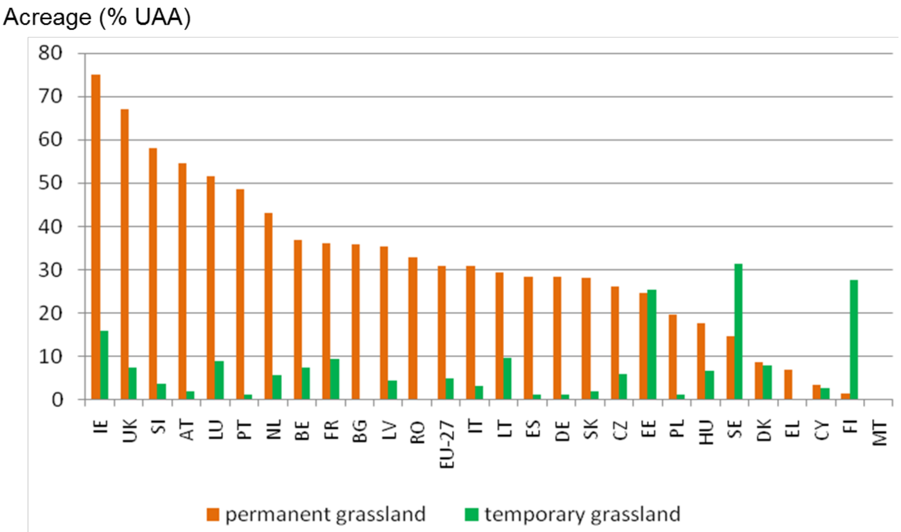

Figure 1. Share of permanent and temporary grassland in EU-27 (adapted from De Vliegher and van Gils, 2010; Eurostat, 2009).

countries, their importance is very low (Czech Republic 0.2%, Hungary 0.2%, Romania 1.4%, Poland 2.0%, Bulgaria 2.2%, Slovakia 2.5%, Slovenia 4.5%).

Maize for silage is an important crop and occupies 5.3 millions of hectares of 3.0% of the UAA. About 58% of the total area is growing in Germany and France. Silage maize is very important in relation to the UAA in Belgium (13.1% of UAA), the Netherlands (12.5% of UAA), Luxembourg (9.9% of UAA), Germany (9.7% of UAA). It represents more than 20% of the grassland area in four countries (Belgium, Netherlands, Denmark and Germany) and in the west part of France and between 10 to 20% in Luxembourg, France, Poland and Slovakia. Maize is used mainly for forage, but Germany uses maize silage on a large scale to generate biogas (De Vliegher and van Gils., 2010).

Evolution of grassland area in Europe and relation with the intensification of the dairy sector

The European grassland area has been significantly reduced during the last 30 years although the estimations of losses of the permanent grassland area vary according to the sources of information (Eurostat, FAO, national statistics). According to Eurostat, in the EU-6, these losses are estimated at about 7.1 million ha between 1967 and 2007 (Eurostat) i.e. about 30% of the value recorded in 1967. This decline is underestimated since the reunification of Germany added about 1 million ha to the total in 1990/91. The losses were very important in Belgium, France, Germany, Italy and the Netherlands (more than 30% decrease) whereas surfaces remained almost stable in Luxemburg (5% increase between 1967 and 2007) and the United Kingdom (4% decrease) (Eurostat database). In Portugal, the surface increased regularly from 0.8 to 1.8 million ha since 1993. In Spain, the surface increased suddenly between 1987 and 1990 and then remained almost stable till 2007. In Bulgaria, the surface was almost multiplied by 3 between 2005 and 2007. These three important increases are probably due, at least partly, to changes in the method of data recording. After 1989, many agricultural areas and especially grassland areas were abandoned in East European countries in transition. It is estimated that at least 30% of grassland areas were abandoned. Marginal grasslands and rangelands tended to be abandoned, especially in mountain and Mediterranean area.

The decline of the permanent grassland area is due to urbanization, afforestation and conversion to arable land. The proportion of permanent grassland in UAA declined from 40 to 29% in EU-6 with similar tendencies in France (40 to 29%), the Netherlands (58 to 43%) and Belgium (48 to 37%). According to Eurostat, the decline of the permanent grassland area seems to be reduced or even stopped after the CAP reform of 2003 which introduced conditionality linked to the maintenance of permanent grassland area to the payment aid. However this trend is not so clear in the FAOSTAT database. Indeed, national statistics reflected aggregated data that can mask contrasting regional evolutions. For example in France, this measure has not prevented a sharp decrease in permanent grassland areas in Lower Normandy and Pays de la Loire (15% or more). The destruction of permanent grassland was also important in the northwest of Germany in favor of maize crop.

The area of poor permanent grassland (rangeland) has marginally decreased from 13.2 to 11.5 million ha between 1990 and 2007 for eight countries (Belgium, Denmark, France, Ireland, Luxemburg, Spain, Netherlands, United-Kingdom). Also the total temporary grassland area can be considered as stable between 2001 and 2007.

The dairy cow population was almost stable between 1975 and 1983 but it felt down by 10 million heads in the EU-9 between 1975 and 2007 (decrease of 40% of the population of 1975 from 25.0 to 15.4 million) after the implementation of the milk quotas in 1984. This evolution was observed for a fixed amount of milk produced thus indicating a very significant increase of milk production per head. At the opposite, the suckling cow population increased by about 3 million head between 1975 and 2007 but the total number of bovine heads decreased by almost 7 million heads. The evolutions are different among countries. The dairy cow population declined sharply (about 50%) in Belgium, Denmark, France and Luxemburg. The substitution with suckling cows was though almost total or even more in Belgium, Greece, Ireland, Luxemburg and Portugal whereas about the half of the dairy cow population was replaced in Germany and France. In Italy and the United Kingdom, the suckler cow population declined, though the dairy cow population also declined, respectively by 35 and 40%.

Finally the reduction of grassland area has been tightly correlated to the reduction of the total number of cows (Figure 2) which in turn is a consequence of the increase of milk yield per cow in a context of milk quota. The intensification of dairy production had led to the development of annual crop and green maize for silage and the utilization of cereals and soybean meal at the expense of permanent grassland. Because part of the concentrate can be used as human food (cereals, soy protein…) whereas grasslands are not edible, one may wonder whether these changes have increased the net contribution of ruminant livestock to the protein security.

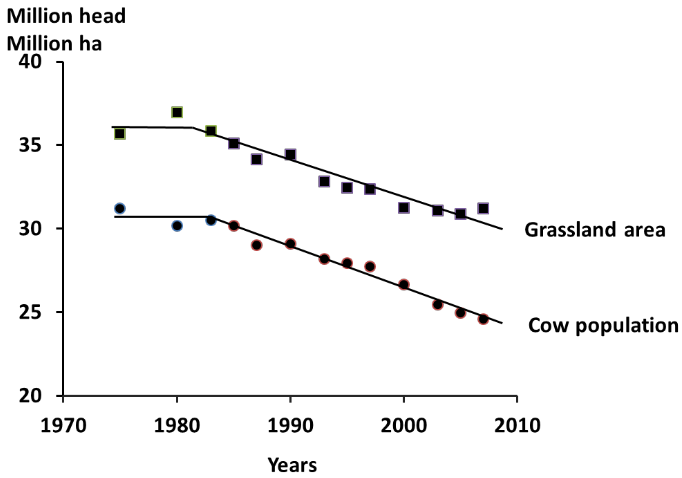

Figure 2. Parallel evolution of permanent grassland area and cow (total dairy and suckler) population in EU-9 (Eurostat, 2010; own calculations).

Contribution of grassland based production system to food security

The overall nitrogen (protein) efficiency of an animal is usually the ratio of nitrogen (proteins) outputs in products and input from ingested nitrogen (protein). In the case of young growing animals, the nitrogen retention rate is also mentioned. This ratio is always far lower than 1.0 and the remainder part of ingested nitrogen is excreted in urine and faeces. The efficiency can also be expressed by the reverse ratio that is the amount of plant protein consumed per kg of animal protein which reflects more directly the competition for the plant resources between the feed for animals and food for humans.

The apparent low protein efficiency of ruminants

Data on livestock nitrogen (protein) efficiency were synthesized by Peyraud *et al.* (2014a). Literature efficiency data show that ruminants are far less efficient than monogastric animals. The efficiency is minimal for dry adult cow or sheep, varies from 10% (dairy heifers) to 20% in growing and finishing animals and is higher in lactating dairy cows (25 to 30%). It takes more than 3 kg of plant protein to produce one kg of milk protein and between 5 and 10 kg of plant protein to produce one kg of bovine proteins. In comparison, a fattening pig fed diets based on cereals and soybean meal, retains about 30-35% of the nitrogen meaning that it takes on average 3 kg of protein of plant protein to produce 1 kg of animal protein. The protein efficiency is of the same order of magnitude for egg production. It is higher for broilers (40% and even 45% for the most productive strains). It takes 2.2 kg of plant protein to make 1 kg of broiler protein on average. These differences are explained primarily by the fact that ruminants are fed with forage-rich diets which are less digestible than diets fed to monogastrics animals.

Advances in genetic merit increased animal productivity and has led to a significant and continuous increase in nitrogen efficiency in all species. The more a dairy cow produces, the highest is the nitrogen efficiency. Efficiency increases by about 5% per tonne of milk on average. Suckler (beef) cows are less efficient than dairy cows, in particular because of their low milk production. Efficiency also increases when the nitrogen (protein) content of the diet is reduced. In dairy cows, efficiency is the highest (about 30%) for maize silage based diets supplemented with soybean meal and can be less than 25% for diets based on green forages with a high protein content (Peyraud *et al.*, 1995). This is why a large number of publications have proposed reducing the proportion of pasture grass in ruminant feeds in favour to a maize silage diet (feed depleted in protein) and a supplementation with soybean concentrates (van Vuuren and Meijs 1987; Valk 1994).

These conclusions suggest that individual cow production has to be increased to be more efficient especially as the same reasoning also applies to the reduction of methane emissions per liter of milk (FAO, 2006; Steinfeld *et al.*, 2006). However this reasoning forgets that the increase in milk and meat production per ruminant and time unit (milk per lactation, average daily live-weight gain) is obtained through an increased consumption of concentrates. These concentrates contain a large proportion of protein that can be consumed by humans. Their use in livestock feeding increases the competition between feed for ruminant and food for humans whereas ruminant have the unique capability of using grassland to produce protein of high nutritional value.

A new insight to the contribution of ruminant grassland based production system to global food security

Competition between feed and food does in fact concern those proteins of plant origin that are consumable by human but are actually consumed by animals. The amount of edible protein of animal origin produced per kg of human edible protein of plant origin is an unbiased view of the contribution of livestock to food security. If the ratio is greater than 1.0, the animal production system positively contributes to food security, if the ratio is below 1.0, the animal production system consumes more plant edible protein than it produces animal protein and if the ratio is 1.0, the system is neutral from the food security point of view. This ratio does not consider the higher nutritional value of protein of animal origin. Taking into account this difference, a ratio of 0.8 would be sufficient to at least maintain a neutral contribution since human beings must ingest less animal protein than plant protein to meet their protein requirements.

The proportions of edible protein in plants are variable between authors (Table 1). Indeed these proportions can be highly variable depending on technological processes and cultural or culinary traditions. In addition, process may change over time. Ertl *et al.* (2015a) expressed the edible fraction of various crops according to three scenarios (Table 1): a low scenario corresponding to a low usage, an average scenario which corresponds to the average data of the literature and a high scenario corresponding to the highest value of literature and potentially attainable values with innovative technologies or changes in eating habits (increased consumption of whole grains, for example). Fresh herbage and grain by-products (wheat bran, gluten feed, distillers grains ...) and beet or citrus pulp do not contain edible protein or very low amounts (i.e. up to 20% for wheat bran). The content of edible protein is high for grains such as wheat, maize and peas (i.e. 70%) and is intermediate for barley (i.e. 60%). Some protein

Table 1. Proportion (%) of edible protein in various feedstuffs.

	Wilkinson (2011)	Ertl *et al.* (2015a)[1]		
		Low	Medium	High
Grass (fresh, conserved)	0	0	0	0
Maize silage	0	19	29	45
Wheat	80	60	80	100
Wheat bran	20	0	10	20
Beet pulps	20	0	0	0
Peas	80	70	80	90
Rapeseed cake	20	30	59	87
Soybean cake	80	50	71	92

[1] A low scenario corresponds to a low usage, an average scenario corresponds to the average data of the literature and a high scenario corresponds to the highest value of literature and potentially attainable values with innovative technologies or changes in eating habits.

from cakes could potentially be extracted for the production of concentrates and the remaining part of protein could be isolated for human diet. The edible protein content of soybean meal ranges from 50 to 90% and lower values were reported for rapeseed cake (0 to 80%). These data show that the lower is the proportion of forage in the diet the lower is the contribution of ruminant to the production of protein.

Feeding animals mainly from non-edible resources can be seen as a conceptually interesting issue from a global food security point of view. Data from Ertl *et al.* (2015b) clearly show that it is possible to produce milk from grassland and crop by-products without using starch. The contribution of various EU livestock production systems to food security was recently evaluated.

In France, the efficiency of different dairy systems were recently analysed using data of the national survey of dairy farms (Institut de l'élevage, 2015) and the three scenarios proposed by Ertl *et al.* (2015a) for the edible fraction of various protein of plant origin (Laisse, unpublished, Table 2). In the lowland, grassland-based dairy systems have a very positive contribution to the supply of edible protein regardless to the selected scenario and more intensive systems using a lot of maize silage and concentrate containing a high proportion of edible protein are far less efficient, contrary to the conclusions that could be drawn from an analysis of the total plant protein consumption (see above). These intensive systems are neutral for the low scenario or consume more edible protein than they produce for the high scenario. Similar results were reported by Ertl *et al.* (2015a) from 30 Austrian mountain dairy farms where the protein efficiency is negatively correlated to the amount of concentrate distributed per kg of milk ($r = -0.82$). The efficiency of dairy system is lower in mountain than in the lowland because cows are fed with more concentrate per litre of milk to compensate for long winter period and medium quality forages. Finally, results heavily depend on the assumptions made for the proportion of edible protein in plant material thus underlying the necessity to determine more precisely these proportions in various countries and technologies.

According to Wilkinson (2011), who considered English livestock systems, the grassland-based beef production is broadly neutral (efficiency of 0.95 based on the weight of carcass) while intensive beef production systems using large amount of concentrates have a negative contribution to protein security (efficiency of 0.3). Production systems of monogastric animals are intermediate with an efficiency of 0.47 for broilers and pig and 0.38 to 0.43 for eggs.

Table 2. Evaluation of the contribution of various dairy systems to the net production of protein (Data Institut de l'Elevage 2015, treatment S. Laisse, unpublished).

Dairy system	Milk (kg cow^{-1})	Scenario (kg animal protein per kg edible plant protein)[1]		
		Low	Medium	High
Milk from lowland regions				
>30% maize silage in forage area	8,200	1.05	0.69	0.50
10 to 30% maize silage in forage area	7,300	1.36	0.89	0.65
<10% maize silage in forage area	6,000	2.64	1.82	1.39
Milk from humid mountain regions				
>10% maize silage in forage area	7,450	1.14	0.75	0.55
<10% maize silage in forage area	6,200	2.09	1.43	1.10

[1] A low scenario corresponds to a low usage, an average scenario corresponds to the average data of the literature and a high scenario corresponds to the highest value of literature and potentially attainable values with innovative technologies or changes in eating habits.

The grassland based dairy system are the most efficient systems to produce protein. In the study of Wilkinson (2011), dairy systems produce up to 1.4 kg of milk protein per kg of edible protein of plant origin and are the most efficient on average because they always value a lot of forages which are not edible while monogastric animals always need high quality foods. The National Farm Survey data in Ireland (Hennessy and Moran, 2014) shows the average Irish dairy farm reaches an efficiency of 1.5 kg of milk protein per kg of ingested edible plant protein. Under experimental conditions with very efficient grassland-based dairy systems a record efficiency of 4.8 was reported (Coleman *et al.*, 2010). These data clearly demonstrate the potential of grassland-based dairy cows systems to contribute to sustainable protein production.

The poorer is the ration fed to ruminants the more efficient they are and the more they contribute to the supply of protein to local populations. For example, in Egypt and Kenya, the rations for dairy and beef cattle are mainly based on low quality forages and ruminants also have there a decisive contribution to protein security; efficiency actually tending to infinity because these animals eat virtually no edible proteins (Bradford *et al.*, 1999). FAO (2011) also reported efficiencies of around 20 for farms in Kenya and Ethiopia. In Australia, under very extensive farming conditions, Wiedemann *et al.* (2015) showed that sheep and cattle grazing rangelands produced respectively 7.9 and 2.9 kg of boneless meat protein per kg of ingested edible protein of plant origin while finishing systems based on diets with a high proportion of concentrate only produced 0.5 and 0.3 kg animal protein kg^{-1} respectively for sheep and cattle.

Ruminant are often blamed for the emission of methane and it is often proposed to shift from ruminant to more monogastrics production in order to reduce the C footprint of our diet. But this evolution will to a certain extent increase feed-food competition. All the available data show that, contrary to what is often said, ruminants are very efficient animals to produce proteins provided they are fed with forages. On the contrary feeding ruminant with high amount of concentrate do not appears as an efficient strategy for a protein production point of view.

The challenge of efficient land use for increasing the production of edible protein

De Vries and De Boer (2010) calculated the total area required for the production of animal products using life-cycle assessment in a comprehensive study. In conventional systems, the mean values ranged from 5 to 6.5 m^2 to produce one kg of chicken or pork (i.e. 180 to 220 kg of meat protein per ha), 4.5 to 6 m^2 for one kg of egg (i.e. 210 to 280 kg of egg protein per ha), 1.2 to 1.5 m^2 per litre of milk (i.e. 200

to 250 kg milk protein ha^{-1}) and 20-25 m² kg^{-1} live weight of cattle at fattening (i.e. 30 to 80 kg of meat protein ha^{-1}). Again considering land use efficiency ruminants appears less efficient than pig or poultry.

In the case of ruminants, previous calculations took into account the entire land area, including those on which it is not possible to produce annual crops while precisely ruminants can contribute to food security by using permanent grassland area that cannot be used for food production or lead to very low yields of annual crops. In this case, they are not in competition with the production of edible protein of plant origins. At the global level, 3.35 billion ha are used in this way (Sere and Steinfeld, 1996) by more than 360 million cattle and 600 million small ruminants and provide 25% of animal products. In European conditions, permanent grassland and rangelands cover 73 million hectares or 40% of the utilized agricultural area (Eurostat, 2009).

In fertile plains, where grassland can often be converted to arable land, the question of the relative yield of protein by ruminants on grassland or by crops can be raised considering that crop production leads to a much higher production of edible protein per unit of land than milk or meat production because it by-passes the transformation step by animals. Conversely the reduction of grassland area also reduces the provision of ecosystems services. Precise estimates of land area required for livestock production according to the production system were the subject of few studies. The French national survey of dairy farms shows that milk production averages 6,000 kg milk ha^{-1} of forage area in grassland systems representing 180 kg of protein ha^{-1} (Institut de l'Elevage, 2015). This productivity can be considered as rather low as it requires the contribution of the equivalent of 0,015 hectares of cereals to cover the needs of the herd, but in this situation cows produce more than 2 kg of milk protein per kg of edible protein of plant origin and contributes to maintain one ha of grassland and ecosystem services it provides. In Ireland, dairy systems are designed to maximize milk yield per hectare. According to the National Farm Survey (Hennessy and Moran, 2014), the average Irish dairy farm produces 375 kg of milk protein ha^{-1} of grass and milk protein yield reaches a record of 550 kg ha^{-1} under experimental conditions using optimized grassland management and suitable animal genetic for grassland-based systems.

In comparison, one ha of wheat producing 8 t DM with 12% protein or 1 ha of pea producing 3 t DM with 22% protein respectively produce 780 and 540 kg of edible protein (20% of their protein are not edible). But, these differences in productivity must also be balanced considering the nutritional value of protein produced.

The nutritional quality of food proteins is evaluated by the index digestible indispensable amino acid score (DIAAS) which was proposed by FAO (2013). This index is calculated from the composition of Indispensable Amino Acid and digestibility in the small intestine of each IAA. Proteins of animal origin are characterized by a much higher DIAAS index than the protein of plant origin. The AAI composition of meat and dairy products corresponds to human needs as they were estimated by AFSSA (2007) that is not the case for protein of plant origin and intestinal digestibility of animal protein is higher than that of plant protein (Gaudichon *et al.*, 2002). The DIAAS value average 135 for meat and milk. Among plant protein, soy proteins are the most similar to animal protein with a DIAAS index equal to 102. A mixture of wheat and peas achieves a DIAAS value of 105 whereas pea and wheat have far lower value due to an imbalanced profile of IAA (80 and 60 respectively). This means that it is necessary to eat 20 to 25% more protein of plant origin than protein of animal origin to cover daily human requirements. This nutritional factor must be taken into account when comparing the productivity of land used for producing animal protein versus edible plant protein.

These data demonstrate the potential of grassland-based dairy cows systems to contribute to sustainable protein production. This requires developing more efficient dairy systems based on grazing. This necessitates many levers and technical innovations as increasing stocking rate, extending the grazing season, using multispecies sward and adapted fertilisation, improving herbage quality, using appropriate animals etc. that were described in many publications (e.g. Peyraud *et al.*, 2004; 2010).

Does intensification of dairy systems allow increasing the net protein production?

The increase in milk and meat production per animal and time unit is obtained through an increased consumption of concentrates containing large proportions of edible protein which ultimately leads to reducing the contribution of ruminant to the net supply of protein. The intensification of dairy farming should be analysed regarding its interest for the supply of edible protein although this strategy allows increasing animal protein production per hectare of the farm it relies on virtual hectares of imported feed. For example in France, intensive dairy systems based on maize silage produce more milk protein per hectare than more extensive systems (270 vs 180 kg protein ha^{-1}; Institut de l'élevage, 2015), allow to produce crops on available land not used for grassland production but, in the same time these systems require more imported soybeans and grain for feeding the herd.

For analysing the impact on the real protein efficiency, we simulated a dairy farm (75 ha UAA, 400,000 l of milk or 12,000 kg of milk protein) in an intensive system with maize silage versus a grassland-based system with low inputs of concentrate. For the same level of milk production, the intensive system can use part of the land area for producing annual crops. The intensive system produces more milk proteins per ha of forage (261 vs 166 kg), a little less meat protein because the herd has fewer cows but produces more protein crops. However the intensive system requires buying more soy protein and a little more grain to feed the herd compared to the grassland-based system. In the end, the net production of edible protein hardly differs between the two systems, but the maize based system is much less efficient than the grassland-based system (respectively 0.92 and 1.97 kg of animal protein kg^{-1} of edible plant protein consumed by the herd). The net protein production of these two systems is in fact strongly influenced by the proportion of edible protein in soybean cake. If this latter increases from 50 (as it is stated in Table 3) to 70% (due to technological progresses for example) the net production of edible protein will become very low in intensive systems (206 kg) and will be maintained at a higher level (i.e. 7,700 kg) in the grassland based system.

Do ruminant grassland-based production systems allow high incomes for farmers?

The comparisons made at the world level show that dairying systems maximising grassland utilisation appear to be highly competitive compared to intensive systems based on indoors feeding and concentrates. A study of international competitiveness (Dillon *et al.*, 2008) has shown that the total cost of production is negatively related to the proportion of grass in the cow's diet. This cost is therefore 50 to 60% higher in Denmark and the Netherlands than in Ireland, whereas France and UK are intermediate. However this is a global approach comparing countries where climatic conditions are very different. Ireland benefits from a climate ideally suited for grassland-based production systems but which does not allow producing cereals in a competitive way. To (re)develop ruminant production systems based on grassland in region where farmers can choose between grassland and annual crops, it is worth checking that these systems provide a sufficient income for farmers. Few studies have been published comparing economic performances across farm according to the production system in a fixed regional context. They show that alternative paths to scale enlargement and spurred intensification are feasible.

In France, Peyraud *et al.* (2014) compared average data of grassland-based and more intensive dairy farms from the 'Sustainable Agricultural Network' (SAN) (about forty farms) and from the French Farm Accounting Agency (RICA) between 2008 and 2012. The farms of the French SAN network are on average smaller than those of the RICA network (56 vs 78 ha), use more grass (87 vs 67% of their

Table 3. Simulation of production of edible protein for two contrasting dairy systems (Delaby and Peyraud, unpublished).

Farm characteristics	Maize-based system	Grassland-based system
Land: Grassland – Maize silage – Crops (ha)	12.9 – 35.5 – 26.6	72.1 – 0.0 – 2.9
Yield: Grassland – Maize silage – Crops (t DM[1] ha^{-1})	7.0 -12.0 – 7.0	8.0 – 0.0 – 6.0
Dairy cows (Total livestock units including heifers)	50 (83,3)	63 (98,9)
Stocking rate (Lu ha^{-1} forage area)	1.72	1.37
Milk (kg cow^{-1} $year^{-1}$)	8,700	6,900
Milk (kg ha^{-1} forage area)	8,264	5,547
Total production of edible protein (farm gate)		
Milk	12,000	12,000
Meat (culled cow and calves)	930	1,163
Crops[2]	17,900	2,088
Concentrate required to feed the herd		
Purchases soybean meal (t $year^{-1}$)	77.0	9.1
Purchased or home grown cereals (t $year^{-1}$)	49.5	46.8
Edible Proteins of plant origin[3] required to feed the herd		
as soybean meal (kg $year^{-1}$)	18,400	2,184
as cereals (kg $year^{-1}$)	4,752	4,492
Net production of edible proteins[4] (kg)	7,678	8,575
Efficiency of protein production of animal origin[5]	0.92	1.97

[1] Dry matter.

[2] Assuming that 20% of cereal protein are non-edible.

[3] Assuming that 50% of soybean protein and 20% of cereal proteins are not edible.

[4] Difference between total production of protein at farm gate and consumption of edible protein of plant origin by the herd.

[5] Kg of protein of animal origin per kg edible protein consumed by the herd.

Main Forage Area) and thus less silage maize (11 vs 32%) and produce less cereals (8 vs 20 ha). In spite of a lower quota (266,500 vs 349,900 l yr^{-1}) and a smaller total value of products per agricultural working unit (AWU) (88,454 vs 104,840 € AWU^{-1}), the farms of the SAN network produce an income before tax that is higher (21,907 vs 17,261 € AWU^{-1}) than on the average farms of the RICA, because of savings on the production costs (248 vs 568 € ha^{-1}). These savings relate mainly to the purchases of concentrated feed (154 vs 320 € ha^{-1}) and inorganic fertilizers (21 vs 92 € ha^{-1}). The economic result before tax and without subsidies, which reveals the real technical performance of the system, is much higher in the farms of the SAN network (7,180 vs 1,490 € AWU^{-1}) (Peyraud *et al.*, 2014).

Another study (Samson *et al.*, 2012) has compared the technical and economic performances of dairy farms from three French lowland regions (Brittany, Lower Normandy and Loire Region) according to their intensification level, in a sample of specialized dairy farms from the RICA network over 3 years (2004-2006). Their farm typology distinguishes three classes of intensification/ self-sufficiency rate on the basis of thresholds of input costs: extensive/more self-sufficient (<390 € ha^{-1}), intermediate (between 390 and 590 € ha^{-1}) and intensive/less self-sufficient (>590 € ha^{-1}). The French studies put figures on different farming paths and compare them in terms of performance and viability The three classes of intensification/self-sufficiency based on the input costs per ha of the RICA network are closely associated to a variation of grassland in the main fodder area (grasslands + other green forage cropped on arable area). More self-sufficient farms include more grasslands than less self-sufficient ones. The degree of intensification does not seem to be a key explanatory factor for the differences in technical-economic performances. The differences in net margins per worker between the three levels of intensification are low, Brittany being the only region where the net margin increases with the levels of intensification (respectively 9,800, 10,800 and 12,100 € AWU^{-1} from extensive to intensive levels) whereas, in the other regions, the most extensive class has on average better performances than the most intensive class (respectively 13,600, 10,300 and 6,800 € UWA^{-1} on average). In this study, the most self-sufficient, which are also the more grassland-based systems, appear to be more resilient to price crises because the share of variable costs in the cost of milk production is always significantly lower than in the more intensive systems (0.10 vs 0.13 vs 0.16 € l^{-1} respectively for the extensive, intermediate and more intensive systems) whereas the market price of milk practically does not vary from one system to another. The strong reduction in milk price in 2009 had relatively less impact on the systems of the SAN network than on the specialized farms of the RICA network. The average level of income before the price crisis was reached again in 2010 after an improvement in the milk price level. In the latter study, as well as in the previous one, the variability of the results within farm class is very important which shows that progress in margins exist in all these systems.

In the Netherlands, Oostindie *et al.* (2013) studied a sample of 1000 dairy farms collecting precise farm accountancy data for the 2007-2010 period. A group of so-called 'economical farmers' could be distinguished (using farming style analysis). Keeping costs associated with the acquisition of external inputs as low as possible was key in their strategy. The same applies to financial costs: debts were kept at low levels. They showed that in the economical farms the costs for animal feed per dairy cow equalled 393 € cow^{-1} year^{-1} (in 2010). This is far below the level of large-scale intensive farms (560 € cow^{-1} year^{-1}) and of small-scale intensive farms (619 € cow^{-1} year^{-1}). Similar or even larger differences were found for fertilizer use. In years with relatively good milk prices (2007, 2010), the net farm incomes realized within the different styles were similar, even while the size of the large-scale, intensive farms (1,400,000 kg of milk) was far beyond the one of 'economical farmers' (560,000 kg of milk). However, in years with low milk prices (2008, 2009), the income of the latter was far higher than of large-scale, intensive farmers. A part of the large-scale, intensive farms even faced a negative cash flow.

Conclusions

The competition between feed for animal production and food for human is becoming a crucial issue considering the expected increase of human population. In this context, livestock and particularly ruminant production are often blamed for their inefficient use of resources including land and for their methane emission. The contribution of ruminant production systems to protein security cannot simply be evaluated by the ratio between animal protein production and the total amount of proteins of plant origin consumed because ruminants have the ability to produce high nutritional products from grassland which cannot be used directly in human food. Grassland-based dairy system can produce up to 2 kg of animal protein or even more per kg of edible plant protein consumed by cows and thus have a very positive contribution to protein security. The intensification of ruminant production system with the development of maize silage and the utilisation of high amount of concentrate at the expense of grassland has indisputably contributed to increase protein yield per hectare used in Europe but has also increased the imports of protein thus reducing European autonomy. Their contribution to protein security does not exceed those of grassland-based systems and indeed are often lower and they increase the feed-food competition. The true efficiency of these systems might be even weaker in the future when the development of new technologies will allow using more protein of plant origin (i.e. cakes) in human food.

From a feed security point of view, the challenge will be to increase protein production per hectare for grassland-based system. Protein yield per hectare of grassland are quite variable and thus there is quite considerable scope to improve the performances of dairy systems based on grassland. We must consider a better management of forage production and conservation, better management of grazing (Peyraud *et al.*, 2010, 2014) and utilization of more appropriate ruminant phenotype to maximize forage use efficiency and limit the appearance of livestock inefficiencies such as fall in fertility or rearing mortality.

An increased use of grassland for ruminant production could also bring positive responses to societal demand for more natural practices and could contribute to the provision of various ecosystem services. In addition, several studies also demonstrated that the economic performances of grassland-based systems are similar and sometimes higher than those observed in more intensive systems. However, grassland utilization remains dependent on the willingness of farmers that are often reluctant and on the attitude of the other actors of marketing chains.

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Reasons for grasslands to last in Western Brittany: an agrarian diagnosis

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Abstract

In the context of an overall reduction in area under grass for the benefit of forage crops in dairy farming areas, Aven region is marked out as having an increased share of grasslands in the total cultivated area between 2000 and 2010. This study aims to understand the agro-ecological factors and historical and socio-economic contexts that have led to the upholding of grasslands in this region. This work calls for the analysis of farming systems in the region and the role of grasslands. The study highlights the role of historical land management, which explains that the spatial organization of the farms plots is especially adapted for pasture. It also points out the pedoclimatic conditions in the region that allow grass outputs close to those of maize. In addition, the recent economic history promoted dairy systems instead of poultry and swine (livestock systems with little grassland). Finally, local extension services also promoted grasslands to reduce the feeding cost. However, the increase of farm size and the end of the milk quota threaten the grasslands. The end of the quota is a source of tension between farmers and a cooperative. Farmers want to keep their systems with low feeding cost, whereas the cooperative invests in dehydration facilities and is looking for more milk volume. If the cooperative's approach prevails, then farmers should increase either the number of cows or the milk quantity per cow. Both options would lead to an increase of winter crops and maize.

Keywords: grassland, livestock farming systems, agrarian system diagnosis

Introduction

There is currently a renewed interest in grasslands: promoted by agricultural policies (Chatellier *et al.*, 2010) and valued in several quality labels, they can provide dairy herd food autonomy (Peyraud *et al.*, 2009) while helping to maintain ecosystem services (Le Féon, 2010; Sabatier *et al.*, 2015). Despite these advantages, their area in France continues to decline, especially in plains regions (-2% on average between 2000 and 2010), Agreste, 2010). Reasons for this decrease were studied (cereal specialization, climate, development of rations based on maize silage ...) but there are also areas in plains regions where grasslands have been maintained or increased during the last ten years (Couvreur *et al.*, 2016). In a small area in the Aven region (Southwestern Brittany), the share of grassland increased from 41% to 45% between 2000 and 2010 (Agreste, 2010). The purpose of this study was to understand the agronomic, ecological and socio-economic factors contributing to this increase, in order to identify levers which can be used in other areas in order to maintain grassland surfaces. An agrarian diagnosis of the area was thus led, enabling the characterisation of the different production systems and the role of grassland in each of them (Raffray, 2014).

Materials and methods

The agrarian system diagnosis aims to understand the evolution and the present situation of the agriculture in a region. The method is first based on an analysis of landscape and pedo-climatic conditions through landscape interpretation combined with analysis of geologic, topographic and weather maps, in order to delimitate sub-areas with the same agronomical potential. Within each of the three sub-areas identified 21 interviews of retired farmers were then conducted, in order to analyse in-depth the agrarian dynamics and the differentiation of production systems in each zone, taking into account that farmers' decisions are strongly determined by the pedoclimatic conditions they deal with (Cochet and Devienne, 2006; Devienne and Wybrecht, 2002). Finally, technical and economic interviews of 38 active farmers were held in the sub-areas. For each type of production system, identified beforehand through the understanding of the agrarian dynamics, the aim was to understand how and why farmers combine several activities and practices on their farms, taking care of technical and economic constraints, and to assess their techno-economic performances (Devienne and Wybrecht, 2002). Data collected during the interviews allow modelling of inputs and outputs, thus enabling calculation of the net value added and revenue generated.

Results

The climate in the region is oceanic temperate. In the northern part, soils formed on mica schist substratum are wetter than those formed on granite in the southern part. Wheat and maize yields are consequently lower (respectively 55 q ha^{-1} and 10-12 t of dry matter (DM) ha^{-1}, compared to 70 q ha^{-1} and 14-16 t DM ha^{-1}) and grassland outputs higher (8 to 10 t DM ha^{-1} compared to 7-9 t DM ha^{-1}). Historically, the farmers of the area were landowners with plots well grouped around the farm facilities. Until the '90s, farmers managed to buy plots around the farmstead so that dairy farms of the area have now around 35 to 50 ha (and up to 100 ha) close to the farmstead, with small plot size, due to the lack of land consolidation. The Aven region was in the '60s a dairy and canned vegetables production basin. As vegetables are suited to precede grassland in rotations, they have incited farmers to combine both types of production on their farms. In the '80s, farm size increased, quota policy appeared and canned vegetable production decreased. This led to the development of swine and poultry production. However, several crises have affected these productions in Brittany over the last twenty years. Therefore, in the area studied, the number of dairy farms only decreased by 8% between 2000 and 2010 with a total increase in the area they manage of 36%, whereas the number of other livestock farms decreased by 43% with a decrease of the area they managed of 36% (Agreste 2010). Moreover, on the northern part of the studied region, an agronomic advisor has been promoting grassland-based dairy systems for several years. Current dynamics are still oriented towards larger farm size and the dairy sector faces the increase of dairy production due to the end of quota system, aimed at the global market (by constructing a drying tower to produce milk powder). Within the dairy farms we have identified six systems, according to the respective role of grass and maize in animal feeding, the other productions on the farm, the milk production per cow and the farm localization in the area (Table 1). The economic modelling reveals that the production systems which generate the most net added-value per agricultural worker are the ones with the highest grass area per cow, with the highest fodder efficiency, which thus have a lower level of intermediate consumption, and use the least maize.

Discussion

Our results identify several factors explaining the increase of grassland: climate favours grass growth, farm size increases and plot configuration around the farm facilities allow grazing systems even in large

Table 1. Main characteristics of dairy systems.

Criteria	VL1	VL2	VL3	VL4	VL5	VL6
Feeding period without maize	No	Yes	Yes	No	No	Yes
Maize area (% of UAA[1])	35	24	17	25	18	19
Maize autonomy	No	Yes	Yes	Yes	Yes	Yes
Milk yield per cow (l)	7,500	7,000	8,000	8,000	7,500	7,500
Other productions	No	No	Crops and vegetables	Crops	Beef	Swine
Zone	Micaschiste	Micaschiste	Granite	Granite	All	Granite
LU ha^{-1} of grassland[2]	2.1	1.4	1.9	1.8	1.9	1.4
Grassland area (ha) /dairy cow	0.60	1.04	0.79	0.82	1.0	0.90
Net value added $worker^{-1}$ (k€)	17 to 49	48 to 94	62 to 87	41 to 84	46 to 87	33 to 73

[1] UAA: utilized agricultural area.

[2] LU: bovine livestock unit.

dairy farms, the technical farm advisor promotes a grass feeding system and dairy systems have suffered fewer economic crises. If the plot organization of farms cannot be a lever to increase the grassland areas in other regions (due to land ownership and relationships between owner and tenant) and pedoclimatic characteristics are unchangeable, then two options can be identified from our case study to increase or maintain grassland areas at the farm scale. Agricultural crises could favour grassland areas, emphasizing the main role played by agricultural policy and evolution of prices. As demonstrated with the second pillar of the Common Agricultural Policy (CAP) there is room for manoeuvre to promote grass-based systems (Chatellier and Guyomard, 2010). The second lever concerns advising and milk valorisation. Dairy systems with a grass-based feeding system are complex to manage and they need specific advising to be supported (Coquil *et al.*, 2014). Better milk valorisation implies a better milk quality which can be achieved with a grass-based feeding system (Borreani *et al.*, 2013). Several quality labels already include a minimum quantity of grass in the cow regime. This study confirms also that the most economically sustainable systems are the ones taking the most advantage of grasslands. However, in the studied area, the end of milk quota is a source of tension between farmers, who want to keep their low feeding cost systems, and cooperatives, looking for larger volumes of milk. If the cooperative's strategy prevails, farmers should increase the number of cows and/or the milk quantity per cow. But as farm size growth is now based on the acquisition of distant land, both options may lead to an increase of winter crops and maize in the feeding system and a decrease in the role of grassland in production systems.

Conclusions

This study shows the importance of historic and climatic conditions for explaining the share of grassland in a landscape. But it also places stress on the main role of socio-economic context (CAP, production system equilibrium, milk valorisation, advising) which can be considered in other areas in order to promotes grasslands.

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Identification of feeding systems used on dairy herds in northern Spain: influence on milk performance

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Abstract

The current models of dairy cow production in Spanish Atlantic Arc are mainly based on the available arable land surface for grazing and maize crop. However, the diet varies throughout the year due to the annual dynamic change of forage production. So, the crop area cannot be always associated with the feeding system. The aim of this study was to identify the real feeding systems used on dairy farms in Asturias and their influence on milk production and composition. For this purpose, sixteen farms were selected according to the available surface for grazing and maize. A survey about the offered feed and milk production was compiled quarterly from summer 2014 to spring 2015. The ingredients of the rations were analyzed by cluster analysis identifying five real feeding systems based on the main ingredient of the diet: Grazing; Maize silage; Grass silage; Dry forage; and Concentrate. Milk composition was analyzed by ANOVA considering feeding system and season as main factors. The Maize silage system had the highest milk production and the lowest use of concentrate. This system showed the highest proportions of protein, lactose, solids-non-fat and urea in milk.

Keywords: dairy cow, grazing, maize silage, grass silage, dry forages, concentrate

Introduction

Most dairy farms in Asturias, Spain, are specialized in maize silage production because maize silage is up to 90% of the total roughage in lactating cow diets (Borreani *et al.,* 2013). As a consequence, the dairy production is largely dependent upon Usable Agricultural Area (UAA) destined to maize culture. In the Netherlands, the UAA with maize has increased from nil in 1950 to 230,000 ha at present (Van Dijk *et al.*, 2015), mainly due to converting grassland into maize. Arango and Fernández (2011) identified four dairy production systems in the Principality of Asturias (Spain) based on the UAA used for growing maize. However, the ingredients of the dairy cow diet vary considerably throughout the year due to the annual dynamic change of forage production, the annual rotation of maize-Italian ryegrass and the variation of nutrient requirements according to the animal physiological state. Therefore, the crop area cannot always be directly associated with the ration. For this reason, the aim of this study was to identify the actual feeding systems used for dairy cows through participatory monitoring during one year in typical dairy farms as representative of a significant number of dairy farms in the region in terms of size, forage and crops and livestock systems.

Materials and methods

Sixteen dairy farms were selected, four by each management system identified by Arango and Fernández (2011), and monitored during one year, with a quarterly visit between summer 2014 and spring 2015. At each visit, a survey was conducted about the fresh ingredient offered and milk production. The pasture intake of cows in grazing farms was estimated by subtracting from the theoretical daily dry matter intake based on NRC (2001) equation, the DM supplied in the stall according to the information submitted by the farmer. Samples of total mixed rations and ingredients (i.e. herbage, silages, hay, straw, concentrates, etc.) were taken. Milk was sampled from the bulk tank. Feed samples were analyzed for dry matter, ash, crude protein, neutral detergent fibre and acid detergent fibre by NIRS. Milk samples were analyzed for fat, protein, lactose, solids non-fat and urea contents by MilkoScan. A cluster analysis with the ingredients

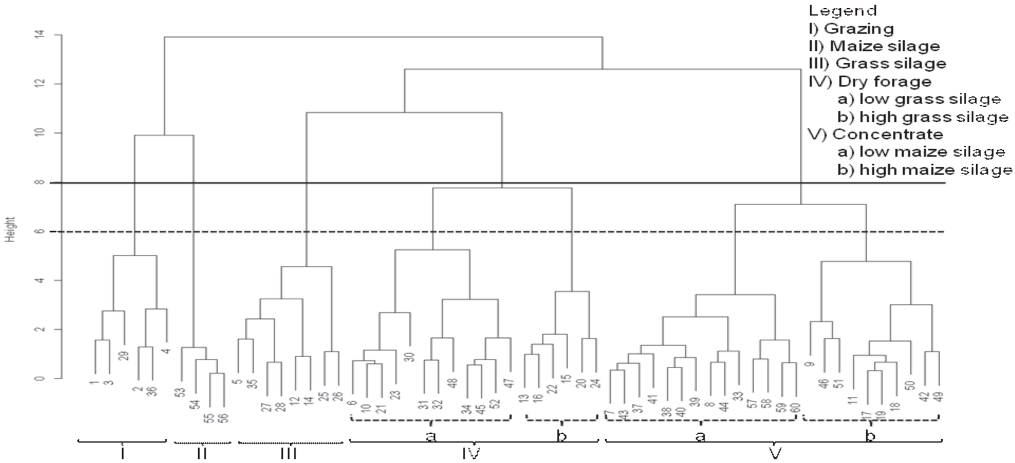

Figure 1. Dendrogram of cluster analysis. Category numbers mean code of ration. The lines represent the threshold similarity value arbitrarily chosen for discriminating seven (dashed line) of five (full line) homogenous clusters.

of the diet in each farm and sampling period was conducted in order to define the characteristics of feeding systems actually used. The squared Euclidean distance was used to calculate the sample similarities. The results of nutritive value of the diet, milk yield and composition were analyzed by ANOVA using feeding system actually used and season as main factors. Duncan test was performed to compare differences between means for groups with different sizes when there were significant differences among feeding systems. The statistical analysis was performed using R package (R Core Team, 2014).

Results and discussion

The cluster analysis identified the feeding systems actually used according to the ingredient offered. The dendrogram generated (Figure 1) shows the possibilities for different groups of rations that can be done according to the cuts at different heights arbitrarily chosen, indicating the similarity as a discriminating threshold to define the homogenous clusters. For discriminating feeding systems, the first cut considered allowed to establish seven clusters, named as: (I) Grazing; (II) Maize silage; (III) Grass silage; (IVa): Dry forage plus low proportion of grass silage; (IVb): Dry forage plus high proportion of grass silage; (Va) High concentrate plus low proportion of maize silage; and (Vb) High concentrate plus high proportion of maize silage. However, further analyses revealed that there were no differences in milk yield and composition between IVa and IVb nor between Va and Vb clusters. Therefore, both sub-clusters were grouped in a second cut and identified as (IV) Dry forage; and (V): Concentrate.

Table 1 shows the ingredient composition and nutritive value of the feeding systems identified, the milk performance and the concentrate needed to produce a litre of milk. The nutritive value meets the nutritive requirements of dairy cows. The average of crude protein was 162±4.88 g kg^{-1} DM with no differences among systems. The net energy was lower in system III than the others feeding systems ($P<0.001$). The highest milk production was obtained in system II ($P<0.001$), while the lowest were in systems I and III. Systems IV and V showed intermediate values. System II used concentrate more efficiently, with 165 g l^{-1} of milk compared to the other groups (374 g l^{-1}, $P<0.001$). System II had the highest proportion of milk protein and solids non-fat ($P<0.001$) than the other systems. Both systems II and V had higher proportions of urea and lactose than the other systems ($P<0.01$).

Table 1. Ingredients composition, nutritive value and milk performance of each feeding system identified: (I) Grazing, (II) Maize silage, (III) Grass silage, (IV) Dry forage, and (V) Concentrate. rsd: residual standard deviation. Values in the same row with different letters differ significantly, ** *P*<0.01; *** *P*<0.001.

	I	II	III	IV	V	rsd	P
Ingredients (g kg^{-1} DM)							
Grass	404		106	101	9		
Maize silage	228	619	20	88	173		
Grass silage	28	73	563	234	259		
Dry forage	12	18	46	233	75		
Concentrate	328	291	266	344	483		
Nutritive value (g kg^{-1} DM)							
Organic Matter	914.3	916.5	915.1	923.1	908.4	38.3	NS
Crude Protein	148.6	163.8	125.5	171.2	169.5	49.0	NS
Neutral Detergent Fibre	389.7a	369.2a	519.5b	444.0a	435.2a	41.1	***
Net Energy (MJ kg^{-1} DM)	6.74b	6.78b	6.03a	6.49b	6.57b	0.260	***
Milk performance (g kg^{-1})							
Yield (kg day^{-1})	14.4a	37.0c	15.3a	23.1b	27.7b	5.68	***
Fat	38.9	38.3	38.6	37.3	38.5	0.23	NS
Protein	31.0a	34.2b	31.3a	31.1a	32.0a	0.13	***
Lactose	47.4a	48.4b	47.3a	47.2a	48.0b	0.08	**
Solids Non-Fat	86.1a	89.8b	85.8a	85.9a	87.6a	0.18	***
Urea (mg l^{-1})	201a	306c	265b	246a,b	285b,c	38.3	**
Concentrate (kg) per litre	0.38b	0.17a	0.44b	0.33b	0.38b	0.102	***

Conclusions

There are five feeding systems on dairy farms in Asturias, defined according to the ingredients on the ration: I) Grazing, II) Maize silage, III) Grass silage, IV) Dry forage, and V) Concentrate. Feeding system affects milk production, milk composition and the concentrate efficiency.

Acknowledgements

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Contract rearing on mountain farms reduces the environmental impact of milk through optimised forage use

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Abstract

Not all regions are equally suited for dairy farming; some areas have more favourable climatic and pedological conditions than others. This is also the case for Switzerland, where lowlands are more favourable than mountains. Hence, Swiss farmers developed a contract rearing system between lowland and mountain farms in order to take advantage of the different regional production conditions. We performed a life cycle assessment of collaborative dairy production using data from four lowland dairy farms that outsourced their young stock to mountain farms and four mountain farms that specialised in heifer rearing (collaborative production). They were compared to four lowland and four mountain dairy farms that kept their young stock on their own farm (non-collaborative production). Collaborative dairy production had a lower non-renewable energy demand and used less mineral phosphorous. In addition, we assessed the farmers' workload. No differences were noted in the lowlands, while in the mountains the workload was significantly lower on farms specialised in heifer rearing. In conclusion, collaboration helped to take advantage of the different production conditions and reduced environmental impacts.

Keywords: division of labour, less favoured regions, dairy, workload

Introduction

In less favoured regions, crop farming is often not viable due to climatic, topographic or pedological conditions. Such regions rely on ruminants that are able to convert grass into valuable products. This is also the case for the area of the Swiss mountains, and therefore dairy farming is one of the activities often found in this region. However, it is difficult for mountain farms to compete with dairy farms from the biophysically more favoured Swiss lowlands. Mountain dairy farms have a lower income (Roesch, 2012), and milk production on mountain farms often causes a higher environmental impact per kg product (Alig *et al.*, 2011). One of the reasons for the disadvantage of mountain farms is that today's high yielding dairy breeds are not well adapted to the steeper and less productive grasslands, and therefore a larger effort is needed in order to fulfil the cows' nutritional needs (Horn *et al.*, 2013). On lowland farms however, the quality of grassland is high, well suited for a grassland based milk production. Farmers from the lowlands sometimes even perceive their grass as being of too high quality to feed less demanding animals, such as heifers needed for restocking. For these reasons, some lowland and mountain farmers started to collaborate in a contract rearing system, where mountain farms rear the heifers, while lowland farms focus on the actual dairy production. We hypothesise that this collaboration is beneficial for the environment, as it optimises the use of available forage resources of both regions, i.e. grassland in the mountains and a combination of grassland and other forages on lowlands. We also hypothesise that it helps farmers to optimise their workload due to specialisation.

Materials and methods

In order to test our hypothesis, we collected data from four collaborating lowland dairy farms outsourcing their young stock to mountain farms and four collaborating mountain farms specialised in heifer rearing, as well as data from four non-collaborating dairy farms in each of the two regions. The non-collaborating farms all rear their young stock on their own land. To characterise the farms, we compared the composition of the feed ration and the milk yield per cow. Farmers' workload was estimated with a workload budgeting tool developed by Stark *et al.* (2014), and compared based on work hours ha^{-1}. The environmental performance was assessed in a life cycle assessment (LCA) with tools developed in the context of the CANTOGETHER project (Teuscher *et al.*, 2014). The functional unit was 1 kg of fat and protein corrected milk (FPCM). The system boundaries included all inputs and processes of the dairy system including the heifer rearing phase. As we had data from four collaborating mountain and four collaborating lowland farms, collaborations between each of those farms were simulated, resulting in sixteen data sets for collaborative dairy production. These were compared to the four non-collaborating dairy farms from both regions, where rearing and milking happened on the same farm.

Differences between groups were tested with a one-tailed Mann-Whitney U test. When interpreting the results, it must be kept in mind that due to the small sample size our farms are not necessarily representative for the corresponding farm types.

Results and discussion

The average share of grass in the feed ration of the dairy cows in collaborating and non-collaborating lowland dairy farms was 62% and 59%, the share of concentrate was 15% and 22%, respectively. On mountain dairy farms, the average share of grass and concentrate in cows' ration was 79% and 12%. Other fodder, such as maize silage or fodder beets, completed the rations. Concentrate use did not differ significantly between each of the farm types. The significantly higher proportion of grass in the feed ration on mountain farms was a consequence of the lower suitability of their land for cropping. This, together with a possibly lower nutritive quality of the grass, also influenced the milk yield of the cows, which tended to be lower on mountain farms ($P = 0.077$). Independently of the region, heifers were fed mostly on grass, either consumed as silage or hay or grazed.

On mountain farms collaboration led to lower workload, while it had no significant effect on the workload of lowland farms. The timesaving on mountain farms created opportunities for off-farm work to increase total income. Two of the four farmers achieved an important part of their family income off-farm.

For most of the environmental impacts, collaborative production and milk production on non-collaborating lowland farms was comparable. For cumulative non-renewable energy demand (nrCED) and P resource use (P use, Figure 1), collaborative milk production had a lower impact. Compared to non-collaborating mountain farms, collaborative production had a lower nrCED, freshwater eutrophication, global warming potential and P use. Non-collaborating mountain and lowland farms showed no significant differences, although there was a trend for higher emissions on mountain farms for most studied impact categories.

These results hint at the environmental disadvantage of dairy production in the mountains already observed by Alig *et al.* (2011). They also show the potential of collaborative production involving both mountain and lowland farms, which could compete with or even outperform dairy production that was purely based in the favoured lowlands. This is in line with findings from a preliminary study based on modelled farms (Marton *et al.*, 2015).

Conclusions

The present study showed promising results for collaboration regarding the environmental impacts and for farmers' workload. The farmers were able to optimise the use of grassland and other feed resources grown on their own farm. The rather extensive heifer rearing is well suited for the grassland based mountain farms and creates opportunities for increased income through off-farm work, while the lowlands offer good conditions for the more demanding dairy cows.

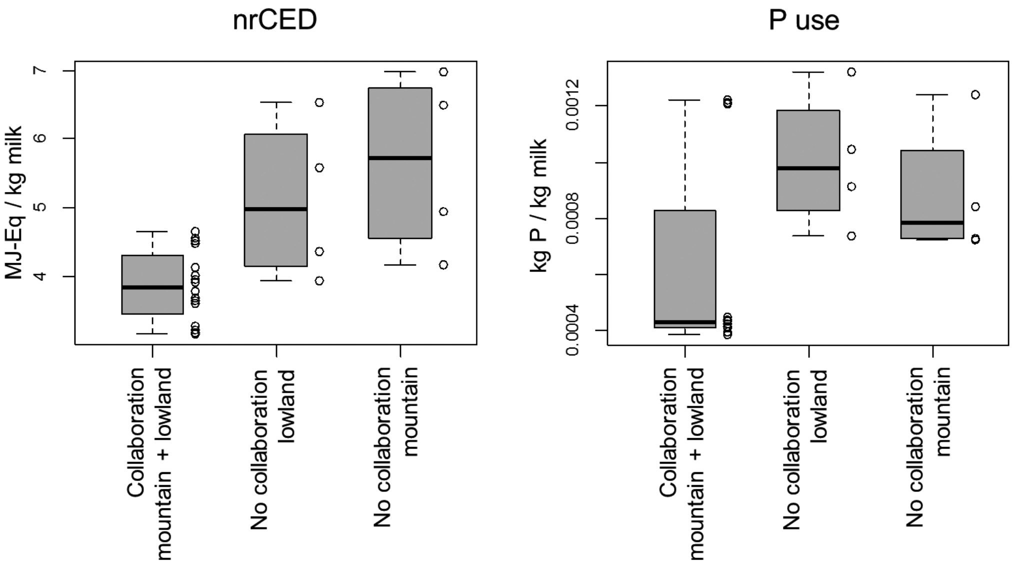

Figure 1. Combined boxplot and strip chart for non-renewable cumulative energy demand (nrCED) and P resource use (P use) per kg fat and protein corrected milk (FPCM) produced under collaboration between mountain and lowland farms, or without collaboration on farms in either the lowlands or the mountains.

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Feeding self-sufficiency levels in dairy cow and goat farms in Western France: current situation and ways of improvement

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Abstract

In the French background, increasing self-sufficiency leads to reduced feeding costs and negative environmental impacts. This paper is focusing on the levels of feeding self-sufficiency on dairy cow and goat farms in Western France. This ratio is defined as the balance between the herd requirements and the resources than can be grown on-farm. It was analysed through three indicators: mass (dry matter (DM)), energy and protein self-sufficiencies. The levels of self-sufficiency in the different production systems and the main solutions to improve them were analysed through national research programs. Dairy cow systems have already a high self-sufficiency in terms of DM and energy. But systems based on maize silage lack proteins. The main focus is then an increased use of grass in all forms and the harvest of protein crops where the climate conditions make it possible to grow them. Dairy goat systems are less self-sufficient. During the last decades, goat farms have gradually turned into intensive farming with more purchased inputs. The very first improvement should be a better consistency between areas and herd size. Other solutions rely on larger resources of forages and on well monitored crop rotations.

Keywords: self-sufficiency, food systems, dairy cow, dairy goat, western France

Introduction

The intensification of livestock production and the development of larger and more specialized farm units have resulted in a decrease in grassland use (Kristensen *et al.*, 2005). Such a model of development based on agricultural intensification weakens the sustainability of farms and jeopardizes their adaptation to global change (Darnhofer *et al.*, 2010; Tichit *et al.*, 2011). 'Agroecology' is a theoretical and conceptual framework developed in order to meet the challenges of global change adaptation of agricultural systems, in order to increase and secure food production (Gliesman, 1998). Agroecology involves: (1) designing farming systems based on biological regulations and interactions between the components of the farm; (2) increasing local feed resources and inputs self-sufficiency; (3) working with local actors (Altieri, 2002). Moreover, feeding self-sufficiency is a major goal for dairy farming, so as to deal with the ups and downs of input costs and of climatic hazards, to improve the traceability of feed and to reduce the dependence of imported proteins.

Feeding self-sufficiency: definitions and methodology

Feeding self-sufficiency is defined as the balance between the herd requirements and the resources that can be grown on farm. Self-sufficiency can be discussed at different scales: the farm, the region, the country. This study focused on Western France (Bretagne, Pays de la Loire and Poitou-Charentes regions) and on both dairy cow and goat farms.

The farms were analysed through three indicators: mass (dry matter), energy (UFL[1]) and protein self-sufficiencies. Information about roughages and concentrates use was provided.An analysis of a French network of pilot farms (Inosys-Réseau d'Elevages) gives us information about self-sufficiency levels in these productions. The aim of this network is to produce, value and then spread technico-economic information on innovative and future milk breeding systems. It is based on 120 dairy farms followed by advisors in Western France.

[1] UFL = Unité Fourragère Lait; Net energy for lactation in MJ = UFL × 6.7 for grass (all types) and UFL × 6.8 for maize silage.

Feeding self-sufficiency levels in dairy cow and goat farms: current situation

Seventy-five dairy cow farms were followed up in 2011 in Western France. Three major production systems were defined: (1) Specialized dairy farms, with more than 30% of forage area composed of maize silage (53% of farms); (2) Specialized dairy farms, with 10 to 30% of maize (32%); (3) Mixed crop-dairy farms, with more than 30% of maize (12%). Table 1 gives an illustration of the cows' diets. Home-grown forages (maize silage and pasture) represent between 64 and 70% of the diet.

Table 2 shows that mass, forage and energy self-sufficiency are high in Western France systems (respectively over 78, 95 and 76%). Variations between feeding systems are low. Links between herd size and fodder area are relevant. Concentrates self-sufficiency is low (from 6 to 24%), with major variations depending on food systems. Mixed crop-dairy farms use their home-grown crops less than do specialized ones. Moreover, protein self-sufficiency is lower, with more variation between the different systems (54 to 68%). The lack of home-grown proteins in the diet is due to the lack of protein concentrates (between 3 and 12% of protein self-sufficiency), in particular in cow systems based on maize silage compared to grass-based ones.

Forty-five goat farms were followed in 2013. According to the database, there is a larger diversity of feeding systems in goat production (Table 1) than in dairy cow production. 50% of the farms have hay-based diets, with 75% of the latter using a majority of legumes (alfalfa or red clover). There is also a large use of maize silage-based diets (31%). 16% of the farms use fresh grass, through pasture or fresh cut pasture (zero grazing). Goat diets are often composed of a mixture between different kinds of roughages, varying according to the season and the available stocks. Concentrates represent 543 kg of DM per goat per year (e.g. 57% of annual goat diet). The dairy goat systems are less self-sufficient than dairy cow systems (Table 2). Fresh grass systems are more self-sufficient (73%), than others (respectively 53% and

Table 1. Food systems in cows and goats farms in Western France (Inosys-Réseau d'Elevages).

	Maize silage(%)	Fresh grass(%)	Hay(%)	Other home-grown roughages(%)	Home-grown concentrates(%)	Purchased concentrates(%)
Specialized dairy farms:						
maize area >30% FA[1]	59	11	1	7	2	20
10< maize area <30% FA	44	20	2	15	4	15
Mixed crop – dairy farms (maize area >30%)	59	10	1	7	2	21
Dairy goat farms	6	3	40	5	10	36

[1.] FA = forage area.

Table 2. Levels of self-sufficiency in dairy cow and goat farms (Inosys-Réseau d'Elevages).

	DM[1] self-sufficiency(%)	Concentrate self-sufficiency(%)	Roughage self-sufficiency(%)	Energy self-sufficiency(%)	Protein self-sufficiency(%)
Specialized dairy farms:					
maize area >30% FA[2]	83	14	98	81	59
10< maize area <30% FA	85	24	96	84	68
Mixed crop – dairy farms (maize area >30% FA)	78	6	95	76	54
Dairy goat farms	61	25	84	56	38

[1] DM = dry matter.

[2] FA= forage area.

57% for hay and silage based diets). Forage share (respectively 64%, 54% and 58%) in the diet is directly linked with self-sufficiency levels.

Feeding self-sufficiency levels on dairy farms: ways of improvement.

The current situation of self-sufficiency differs between dairy cow farms (Rouillé *et al.*, 2014; Brocard *et al.*, 2015) and goat farms. However, ways of improvement are similar. One first improvement is a better consistency between areas and herd size. Other solutions rely on:

- herd management: larger use of grass in the diet, feed efficiency, nature of feed (protein crops, rapeseed meal, by-products);
- crop management: grassland yields and quality of forages (more legumes, multisward), new cultivated feed, well monitored crop-rotation.

In the west of France, two additional research and development schemes for high performances and sustainable goat farming have been created: Patuchev and REDCap (Caillat and Jost, 2015). This organization gives a wide place to (1) experiment in experimental unit and in farms; (2) exchange to support the emergence of innovative research issues and (3) spread of new knowledge on the topics of self-sufficiency and grass use. A national program AUTOSYSEL has been launched by Idele in 2014 with 105 pilot farms and experimental sites to study and show the innovative solutions developed.

Conclusions

Feeding self-sufficiency, in terms of DM and energy levels, is high in French bovine and goat systems of western France. The consistency between herd size and fodder area is good on cow farms, but should be improved on goat farms. The main challenge for dairy farms consists in improving the home-grown feedstuff and protein production, so as to improve protein self-sufficiency. The relatively high availability of land at a moderate price in France is an opportunity to meet feed requirements of the animals with home-grown fodder and crops. This strategy is a major competitiveness issue in terms of production costs for dairy systems and also an efficient solution to reduce losses to the environment, e.g. through nitrate leaching and greenhouse gas emissions. Self-sufficiency is also important in terms of breeding image. More than the question of self-sufficiency in terms of dry matter, energy or proteins, the question of competition between proteins availability for food and feed is important, and puts the notion of edible proteins into the fore.

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Environmental impacts of pasture-based beef production compared to concentrate-based beef fattening in Switzerland

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Abstract

Cattle fattening on pasture is often discussed as a mitigation option to reduce competition between animal and plant production for human nutrition. We performed an life cycle assessment analysis to compare a conventional, concentrate-based beef fattening system with a pasture-based system. Data were obtained from real farms. We calculated 16 environmental impacts, which were analysed related to the animal live weight. Except for deforestation, the pasture-based system exhibited higher impacts for all categories analysed, mainly because of its lower daily weight gain. Targeting the same slaughter weight, the lower daily weight gain of the animals on pasture resulted in a much longer fattening period and a higher need for energy, water and land area as well as higher emissions of methane and nitrogen compounds. In contrast, previous studies on grass based milk production showed that it performed equally well as milk based on feed concentrates. This could be a hint that from an environmental point of view, dairy systems might be preferable over beef fattening systems for drawing value from high-quality grasslands.

Keywords: beef, environmental impact, LCA, roughage, pasture

Introduction

As ruminants are able to convert grass into high value food products, reconnecting cattle to grassland less suitable for crop production might be an option for mitigating the area competition between animal and plant production for human nutrition (de Vries *et al.*, 2015). The majority of the world's agricultural area is covered by grassland (FAOSTAT, 2015) and therefore, grass is a major agricultural resource. In Switzerland, 70% of the agricultural area consists in grassland (BFS, 2014), and Swiss agricultural policy promotes the use of this important domestic resource. Besides, extensive production systems are often perceived by consumers to have a lower environmental impact than intensive ones (Xue *et al.*, 2010). However, the sustainability of grassland-based ruminant systems is controversially discussed (Steinfeld *et al.*, 2006; Gerber *et al.,* 2015). In this study, we assessed the environmental impacts of a conventional, concentrate-based beef fattening system and a pasture-based system in Swiss conditions in order to provide a basis for decision-making and facilitate system-optimisation.

Materials and methods

The environmental impacts of the two systems were assessed by life cycle assessment (LCA) according to SALCA (Nemecek *et al.*, 2010), developed by Agroscope for agricultural systems. The following environmental impacts were examined: non-renewable energy demand, global warming potential, ozone formation potential, demand for phosphorous and potassium resources, land competition, deforestation, water use, eutrophication potential, acidification potential, terrestrial and aquatic ecotoxicity as well as human toxicity potential. System boundaries were set at farm gate; all results were expressed per kg live weight.

Data were obtained from 17 and 13 real conventional and pasture-based beef fattening farms, respectively. Table 1 shows the most important production parameters of the systems analysed.

Table 1. Production parameters of the conventional and pasture-based beef fattening systems.[1]

	Conventional beef fattening	Pasture-based system
Location of farms	Plain region	Plain region
Slaughter age (weeks)	53	90
Slaughter weight (g)	568	543
Daily weight gain (g)	1,279	741
Feed intake per kg weight gain (kg DM kg LW^{-1})	4.8	8.9
Basic ration (kg DM)	1,543	3,326
Pasture grass (%)	–	31
Hay (%)	2	25
Grass silage (%)	12	31
Maize silage (%)	73	13
Sugar beet pulp (%)	13	–
Concentrate intake (kg)	763	94

[1] DM = dry matter; LW = live weight.

Results and discussion

Except for deforestation and resource use potassium, the pasture-based system exhibited higher environmental impacts per kg live weight for all impacts analysed (Figure 1). This was due to the lower digestibility of the roughage as main feed ingredient and the resulting longer fattening period and higher total feed intake. Targeting the same slaughter weight, the pasture-based animals lived 1.7 times longer and needed 1.9 times more dry matter intake per kg weight gain than the conventional animals. This led to higher methane and nitrogen emissions as well as a higher need in energy, land, water and infrastructure.

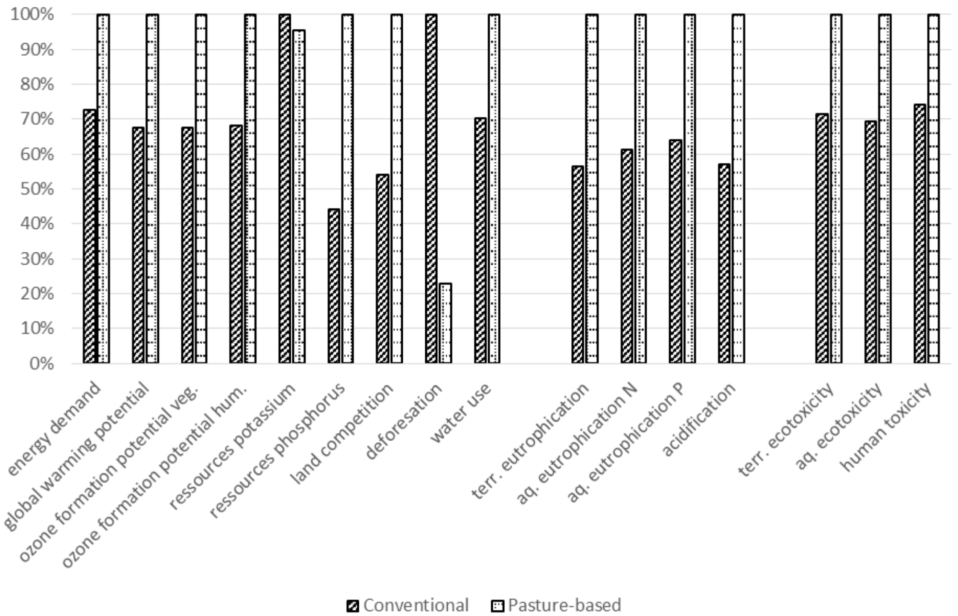

Figure 1. Comparison of the environmental impacts of the conventional and pasture-based beef fattening system. 100% = system with the higher impact.

These results confirm the lower eco-efficiency of grass-based beef fattening systems compared to concentrate-based systems also found in a review study of de Vries *et al.* (2015), although in this study, the results for acidification and eutrophication differed among the studies analysed. Previous studies comparing grass-based and conventional dairy systems were more ambiguous: There was no clear advantage for the one or other system (Alig *et al.,* 2015). With around 30% lower milk yield, the productivity losses in grass-based dairy farming were less pronounced than in grass-based beef fattening (daily weight gain over 40% lower). Therefore, the productivity losses in milk production could be compensated to a higher degree trough the lower inputs for feed production than the losses in beef production. Although the simplifications inherent to LCA calculations lead to notable uncertainties, this could be a hint that in Swiss or similar conditions, dairy systems could be more suitable for grass-based production than beef fattening systems. This is in line with the fact that beef produced from calves bred by dairy cows exhibits lower environmental impacts than beef from calves bred by suckler cows (Alig *et al.*, 2012; de Vries *et al.*, 2015).

Conclusions

Even in a country with high quality grassland and a good grass growth, the pasture-based beef fattening system could not compete with the concentrate-based system in terms of environmental efficiency. Previous studies showed more favourable results for grass-based dairy systems. From an environmental point of view, dairy systems might preferable over beef fattening systems for drawing value from high-quality grasslands.

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The environmental performance of grassland and arable-based dairy farms – a case study from Austria

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Abstract

In Austria 70% of all dairy farms are located in grassland areas. Although grassland-based milk production is dominating, concentrate and N fertiliser are used in varying amounts to increase farm milk output. These agricultural inputs are purchased externally or produced internally on the own arable farm area. In a case study, we analysed how the concentrate and nitrogen input and origin influenced the environmental performance of thirteen grassland-based and nine arable-based dairy farms in Austria. As a main result of the life cycle assessment, the amount and origin of concentrate significantly influenced the environmental impact categories P resource use, land competition, terrestrial ecotoxicity, deforestation and aquatic eutrophication N per kg milk at farm gate. Total and purchased N input correlated positively with the P resource use, deforestation and aquatic eutrophication nitrogen, purchased N input correlated negatively with global warming potential and land competition. Grassland-based dairy farms showed a higher global warming potential and land competition, but a lower P resource use and deforestation potentials per kg milk compared to the arable-based farms.

Keywords: life cycle assessment, agricultural inputs, environmental impacts, milk production

Introduction

On arable land, both the direct production of feed as well as those of feed as silage maize or clover grass is possible. In areas with natural or other specific constraints, an arable use is less viable. Thus, grassland to produce fodder for roughage consuming animals is predominant. These main differences have not only implications on dairy farm management, milk cow feeding and agricultural inputs use, these distinctions have also effects on the environmental impacts of the output. Model studies from O'Brien *et al.* (2012) and Bystricky *et al.* (2014) showed differentiated pictures of the environmental impacts depending on feeding and agricultural resource input. In this paper, we analysed if the theoretical observed differences between grassland and arable based production can be observed in a sample of practical farms.

Materials and methods

Farm management data were obtained from the project 'FarmLife' where in total 22 dairy farms were examined with regard to their environmental performance (Herndl *et al.*, 2016). The farms were classified based on the dry matter intake of dairy cows. If dry matter intake from grass was higher than 80% of the ration, farms were considered as grassland-based (GB, n=13), otherwise as arable-based (AB, n=9). To test the impact of the different agricultural input use to environmental impacts, the parameters concentrate input (home-grown, purchased) and nitrogen input (manure from own animals, purchased – mineral and organic) were used. The environmental performance was analysed using life cycle assessment. The life cycle inventory was computed with the tool FarmLife-Calculate (Herndl *et al.*, 2016), the impact assessment was performed with the software SimaPro v7.3. The system-boundary is limited to the dairy enterprise of the farm and the functional unit is kg milk at farm gate. The attribution of emissions to milk and its coproduct meat was based on economic allocation. The analysed environmental impacts are described in Bystricky *et al.* (2014) and are as follows: land competition, P resource use, deforestation, non-renewable energy demand, global warming potential, aquatic eutrophication nitrogen and phosphorous and terrestrial ecotoxicity. Statistical analyses were conducted using procedures of the SAS program (SAS Institute, 2000). Linear regressions were performed with Excel.

Results and discussion

The average annual milk production of the GB farms was 129,698 kg and of the AB farms 231,365 kg. The total concentrate input, home-grown concentrate, and N purchased were higher in AB farms and N originated from animals (own production) was lower than in GB farms (Figure 1). Significant differences (P<0.05) of the two farm types regarding environmental impacts were observed for land competition, P resource use, deforestation and global warming potential. Similar differences were also observed by a model study of Bystricky *et al.* (2014).

The high scattering of some environmental impacts refers to a possible optimising potential. To identify influencing variables, a linear regression analysis was made. A higher total concentrate input per kg milk led to higher P resource use, deforestation and terrestrial ecotoxitcity per kg milk (Table 1). The same correlation was observed for concentrate purchased, which had in addition a positive correlation with aquatic eutrophication nitrogen. This was also found in the study of and Hörtenhuber *et al.* (2013). Home-grown concentrate showed only a positive correlation to the impact category land competition. A positive linear correlation was found between total N input (mineral fertiliser and farm manure) and P resource use, deforestation and aquatic eutrophication N. Organic nitrogen originating from own animals positively correlated with the non-renewable energy demand whereas N purchased had a positive relation to aquatic eutrophication N.

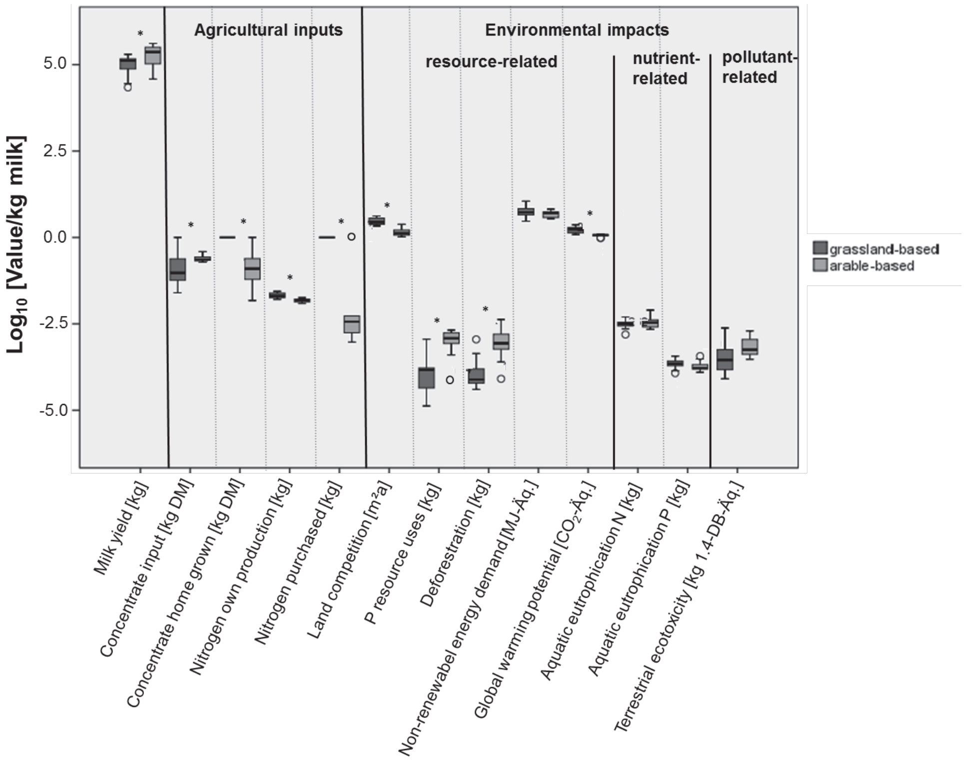

Figure 1. Agricultural inputs and environmental impacts per kg milk at farm gate of grassland and arable-based dairy farms (results are expressed as common logarithm to harmonise the scale. Means are significant different at P<0.05, n=22; outliers are marked with circles).

Table 1. Correlation coefficients between environmental impacts and agricultural inputs per kg milk at farm gate.

Environmental impacts	Milk yield (total)	Concentrate input	Concentrate purchased	Concentrate home-grown	Nitrogen input	Nitrogen urchased	Nitrogen own production
Non-renewable energy demand	-0.47*	+0.03	+0.29	-0.38	+0.26	-0.19	+0.58**
Global warming potential	-0.61**	+0.24	+0.03	-0.46	-0.10	-0.52*	+0.82***
P resource uses	+0.41	+0.66***	+0.62**	+0.23	+0.56**	+0.66***	-0.44*
Land competition	+0.60***	-0.06	+0.02	+0.51*	-0.16	-0.62**	+0.70***
Deforestation	+0.50*	+0.53*	+0.53*	+0.13	+0.71***	+0.89***	+0.44*
Aquatic eutrophication N	+0.49*	+0.38	+0.48*	-0.13	+0.57**	+0.55**	-0.08
Aquatic eutrophication P	-0.20*	+0.03	+0.29	-0.37	+0.24	-0.10	+0.36
Terrestrial ecotoxicity	+0.20	+0.48*	+0.65**	-0.11	+0.41	0.35*	-0.23

* $P<0.05$; ** $P<0.01$; *** $P<0.001$.

Conclusions

There are several differences in farm management between GB and AB dairy farms. These distinctions range from varying milk cow feeding to different agricultural inputs use, however, with effects on the environmental impacts of the output. The analysis performed in this paper showed not only the differences in the environmental performance of the two farm types, it also showed how agricultural inputs influence environmental impacts of the output milk. Concluding, the efficient use of especially purchased concentrate can be an optimising potential for GB farms, whereas an effective use of particularly purchased N can help in lowering the environmental impacts for AB farms.

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The effect of social factors on the extent of grazing

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Abstract

The extent of grazing is affected by technical factors like stocking density per ha grassland. Previous research suggested that farmers themselves also play a key role in determining the extent of grazing since they decide on the day to day management on their farm. However, the effect of social factors on the extent of grazing had not been quantified. The aim of this research was to study the technical and social factors that affect the extent of grazing on commercial dairy farms. Based on the Theory of Planned Behavior, it was hypothesised that the extent of grazing is influenced by the attitude of farmers towards grazing, subjective norms about grazing, perceived behavioural control of grazing and technical possibilities for grazing. An on-line questionnaire was sent to commercial dairy farmers in the Netherlands and 212 valid responses were obtained. Results were analysed using factor analysis and linear regression analysis. Combining technical and social factors in a multiple linear regression model could account for 47% of the variation in the extent of grazing. The results imply that future work on grazing in the Netherlands should take the mind-set of the farmer into account.

Keywords: attitude, dairy, grazing, mind-set of the farmer

Introduction

In the last decade, grazing of cattle has become a societal issue in the Netherlands. The dairy sector wants to support grazing by influencing the factors that are affecting the extent of grazing on farms. In general, grazing is seen as an economically attractive activity (e.g. Dillon *et al.*, 2005; Peyraud *et al.*, 2010). The extent of grazing is obviously depending on a number of technical factors, like available land area for grazing and number of dairy cows present. Changes in those technical factors could lead to changes in the extent of grazing per cow. But technical factors are not the only influencing factors. Farmers play a key role in determining the extent of grazing of their dairy cattle since they decide on the day to day management on their farm. From on-farm participatory research, it is known that personal values, preferences, experiences and habits of farmers are very important in the decision whether to graze or not to graze (e.g. Reijs *et al.*, 2013; Van den Pol-Van Dasselaar *et al.*, 2008). However, this has not been quantified. Think for example of the beliefs of farmers with respect to grazing, e.g. the benefits that farmers expect from grazing or their personal view on capabilities to practise grazing. Therefore, when studying the factors that influence the extent of grazing, the opinions and thoughts of farmers should be included. Their perception on the effect of grazing will definitely influence their decision on whether to graze and, if grazing is chosen, to what extent grazing is practised. If farmers do not perceive positive effects of grazing or if they expect too many barriers to grazing, they will not be motivated to start grazing. The aim of this research was to study the technical and social factors that affect the extent of grazing on commercial dairy farms.

Materials and methods

A conceptual framework was developed based on the Theory of Planned Behavior (Ajzen, 1991), in which the extent of grazing is influenced by:

- the attitude towards grazing (which is a function of the belief of the dairy farmer in the effect of grazing on various topics, for example does the dairy farmer believe that grazing leads to more job satisfaction, and the importance of these topics);
- subjective norms about grazing (which is a function of the social normative beliefs with respect to grazing, for example does the dairy farmer believe that grazing leads to a better image of dairy farming, and the importance of these beliefs);
- perceived behavioural control of grazing (which deals with the perceptions about having the necessary resources available, like knowledge and infrastructure); and
- technical possibilities for grazing (here expressed as milk production per ha).

Based on this framework, an on-line questionnaire with questions on attitude, norms and perceived behavioural control was developed and sent to 1109 dairy farmers. The answers were then combined with technical and economic data from the annual accounts of these dairy farmers. This resulted in a total of 212 valid respondents. The effect of social and technical factors on the extent of grazing was studied in two steps. First, a factor analysis was carried out to understand the structure of the items of attitude, subjective norm and perceived behavioural control. Second, a multiple linear regression analysis was carried out with extent of grazing as dependent variable and attitude towards grazing, subjective norm about grazing, perceived behavioural control of grazing and technical possibilities for grazing as independent variables.

Results and discussion

The main characteristics of the 212 farms are given in Table 1. The mean herd size and the mean annual milk production were near the average of the Netherlands (LEI, 2014).

Factor analysis revealed that attitude of the farmer towards grazing consisted of two components: the component Farm Continuity Beliefs (related to the belief of the farmer with respect to the effect of grazing on income, labour, animal health, mineral losses, job satisfaction and farm development) and the component Grass Yield Beliefs (related to the belief of the farmer with respect to the effect of grazing on grass growth and grass utilisation). Only one component was extracted for subjective norm about grazing (Social Normative Beliefs) and for perceived behavioural control.

There were significant effects of grazing on the different components. On average, Social Norms were seen as a driver for grazing; dairy farmers believed that the outside world supports grazing. Grass Yield was seen as a barrier to grazing; farmers believed that grazing has a negative effect on grass yield. Farmers that grazed, associated grazing with Farm Continuity and Perceived Behavioural Control. Farmers on

Table 1. Characteristics of the 212 dairy farms in this study: total area of forages (grass and maize), total grassland area, herd size, milk production, age of the dairy farmer, number of people working in the farm.

	Mean	Standard deviation	Minimum	Maximum
Forage area (grass and maize) (ha)	45	18	11	157
Grassland area (ha)	37	15	8	111
Herd size (number of dairy cows)	87	38	22	323
Milk production (kg FPCM cow^{-1} yr^{-1})[a]	8,801	968	5,403	11,110
Age of the dairy farmer	52	9	27	93
People working in the farm (FTE)[b]	1.7	0.6	1	4

[a] FPCM = Fat-protein corrected milk.

[b] FTE = full-time equivalent.

the other hand that did not graze had the opposite association. This was consistent with their choices in grazing management.

Combining Farm Continuity Beliefs, Grass Yield Beliefs, Social Normative Beliefs, Perceived Behavioural Control and technical possibilities (expressed as Milk production in kg ha^{-1}) in a multiple linear regression model could account for 47% of the variation in the extent of grazing. Farm Continuity Beliefs, Perceived Behavioural Control and Milk production per ha were significant at the 0.01 level in this model and Social Normative Beliefs was significant at the 0.05 level (Table 2).

Table 2. Results of the multiple linear regression with extent of grazing (hours cow^{-1} yr^{-1}) as dependent variable and Farm Continuity Beliefs, Grass Yield Beliefs, Social Normative Beliefs, Perceived Behavioural Control and Milk production per ha as independent variables.

	B	Std Error	β	Significance
Constant	-410	372	–	0.271
Farm continuity beliefs	426	79	0.332	0.000
Grass yield beliefs	93	57	0.091	0.104
Social normative beliefs	127	60	0.123	0.034
Perceived behavioural control	171	50	0.190	0.001
Milk production per ha	-0.059	0.011	-0.290	0.000

Combining data on the extent of grazing, technical data and data on personal values, capabilities and preferences of dairy farmers had not been done before in a quantitative way and provides a unique first insight into the corresponding relations.

The intended use of the research results is to apply the obtained knowledge and insights in discussions with different stakeholders like farmers and policy makers in order to support grazing. For effective knowledge dissemination on grazing to specific groups especially the knowledge about drivers for grazing and barriers to grazing is important. Potential drivers for grazing can be used to stimulate grazing and actions can be defined to overcome potential barriers.

Conclusions

Combining technical and social factors in a multiple linear regression model could account for 47% of the variation in the extent of grazing. The results imply that future work on grazing in the Netherlands should take the mind-set of the farmer into account.

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How does grazing duration per year affect the environmental impacts of dairy farming?

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Abstract

In today's Europe, we observe two contradicting tendencies: On the one hand, pasture grazing is declining, on the other hand, consumers call for production systems where animal welfare is increased by allowing dairy cows to express their inherent behaviour. At the same time, environmental issues such as climate change remain a matter of importance. Hence, in a study with twenty-two dairy farms we assessed whether high grazing duration per year had an effect on the environmental performance of milk production. Focusing on the environmental burden per ha farmland, dairy farms with a longer grazing duration performed significantly better than those with shorter ones for all assessed categories, e.g. non-renewable energy demand, global warming potential, and terrestrial ecotoxicity. However, when we turned the attention to the assessment per kg milk, the life cycle assessment results for longer grazing duration showed a diverging picture with benefits for ecotoxicity and the use of P resources and drawbacks for global warming potential. Overall, dairy farming with high grazing duration was better or at least similar for many categories in comparison with low grazing duration farms, with some exceptions as global warming potential when assessing per kg milk. For an even better achievement further efforts should be made to decrease global warming potential from grazing cows (e.g. by improving pasture quality and animal genetics, or using feed supplements) in order to reduce the environmental impacts per kg milk without compromising the results per ha farmland. This would help to promote grazing in European dairy farming.

Keywords: grazing duration, life cycle assessment, environmental benefits, farm management, dairy farming

Introduction

In today's Europe, two contradicting tendencies can be observed: On the one hand, pasture grazing is declining as high lactating cows are fed mainly indoors. On the other hand, consumers' awareness of animal welfare issues is increasing, and numerous stakeholders consider grazing an important part of the inherent behaviour of dairy cows. The same consumer group is often also concerned about the environmental performance of products. The objective of this study was to assess how grazing duration affects the environmental performance of dairy production.

Materials and methods

Twenty-two commercial Austrian dairy farms provided agricultural management data for the year 2014. Grazing duration was calculated from the number of pasture days per year multiplied with the average hours spent on the pasture per day. We formed two classes: (1) a high grazing duration (n=10; average grazing duration per year: 3,880 hours) and (2) a low grazing duration group of farms (n=12; average grazing duration per year: 630 hours). As a threshold, we chose 12 hours grazing during 220 days, i.e. 2,640 hours per year. Being above this means these farms let their cows graze the majority of the vegetation period.

In the high grazing duration group on average grazing supplied 37%, dried or ensiled roughages 58%, and concentrates 5% of the feed intake (in dry matter). For the low grazing duration group the feed intake was on average split in 8% from grazing, 78% from dried or ensiled roughages, and 14% from concentrates, respectively.

For the environmental assessment we used the FarmLife-methodology (Herndl *et al.*, 2016), a life cycle assessment (LCA) approach at farm level. The spatial system boundary was set at the farm gate. The temporal system boundary is one calendar year for animal husbandry and permanent grassland. For arable crops, it lasts from the harvest of the previous main crop to the harvest of the current main crop, for temporary grassland from the last cut in the previous year to the last cut, or the ploughing, in the main year.

Agricultural systems have two main functions for society: (1) the preservation of natural livelihood for future generations, and (2) the provision of edible products. Therefore we chose two functional units to reflect these two functions,: (1) 1 ha utilised agricultural area in one year and (2) 1 kg of milk. The statistical tests for all comparisons between the two farm groups were performed by using the tool 'R' applying a one-tailed Mann-Whitney U test. The selection of the assessed environmental impacts is based on SALCA (Nemecek *et al.*, 2010).

Results and discussion

Per ha farmland and year, the high grazing group of farms had significantly lower values for cumulative non-renewable energy demand (nrCED), global warming potential (GWP; Figure 1), both aquatic eutrophication N and P, terrestrial ecotoxicity, use of P and K resources, land competition, land use change, and water use. The lower stocking density in the high grazing group was one reason for these positive results. Another important reason was the use of far fewer concentrated feedstuffs (70% less per cow on high grazing farms compared to low grazing farms). In addition, the greater use of purchased concentrates explained the higher land competition for low grazing farms systems, as they needed more agricultural surfaces off-farm for the purchased feedstuffs. In a world with growing human population and increasing demand for food, this is key, as the competition for land will grow further.

Analysing the LCA results per kg milk showed a diverging picture: For several impact categories such as nrCED, aquatic eutrophication N and P, and water use, there was no difference between the dairy farms with high and low grazing duration. For other impacts such as GWP, land competition, and land use change, the impacts of the high grazing farms were significantly higher than of the low grazing ones (Figure 2). Moreover, there were impact categories, such as terrestrial ecotoxicity or resource use of both P and K, where the group of high grazing farms had a significantly lower impact than the low grazing farms.

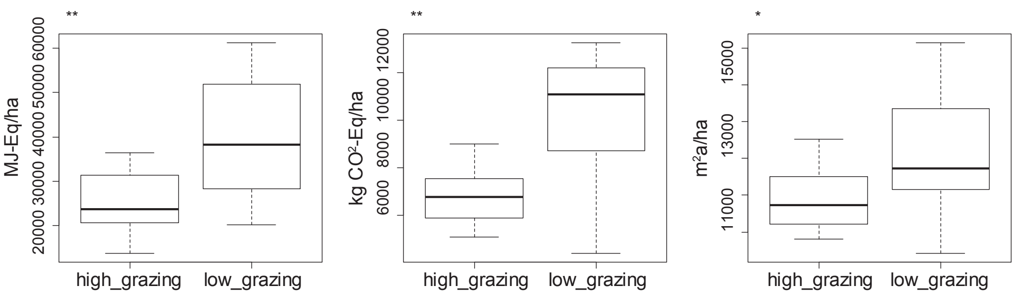

Figure 1. Environmental impacts per ha farmland and year for cumulative non-renewable energy demand, global warming potential, and land competition (from left to right); * = significant different at *P*<0.05; ** = at *P*<0.01; n=22) for the systems high grazing and low grazing duration, respectively.

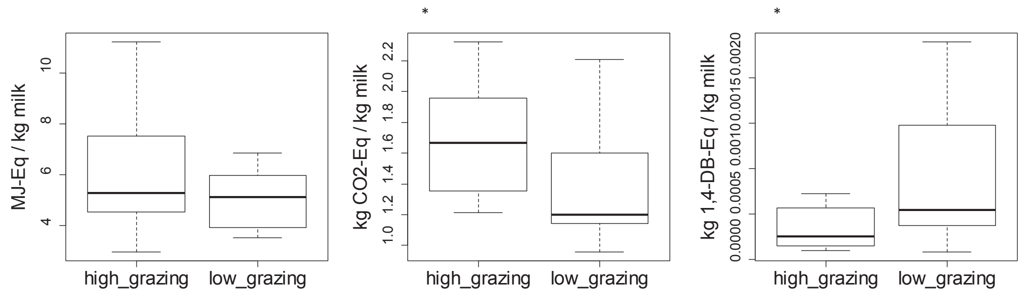

Figure 2. Environmental impacts per kg milk for cumulative non-renewable energy demand, global warming potential, and terrestrial ecotoxicity (from left to right); * = significant different at $P<0.05$; n=22) for the systems high grazing and low grazing duration, respectively.

One of the reasons for these trade-offs per kg produced milk was the milk yield (kg cow^{-1} × a) that on average was 28% lower on high grazing farms. For environmental impacts where the advantages of high grazing duration were small, the reduced milk output resulted in a less favourable LCA result. However, altogether there were few trade-offs, which is encouraging. Need for action exists for reducing the GWP. Consequently, in a further step the assessed farms should be encouraged to apply greenhouse gas mitigation techniques such as feed supplements, better pasture quality and on a longer term improved cow genetics.

Conclusions

Producing milk with high grazing duration had significant or very significant environmental advantages regarding the effects per ha farmland, i.e. the livelihood preservation function. At the same time, we observed for the high grazing farms some negative environmental effects with respect to the productive function, e.g. for GWP. Moreover, such a production strategy is expected of having positive effects on the animal welfare as the cows can pursue their natural behaviour and on biodiversity due to more disturbed and hence structured grasslands offering habitats that are more diverse. These are encouraging results to promote grazing of dairy cows. Being aware of the small sample size located in only one European country, our results call for a larger survey of the hypothesis of the environmental advantages of producing milk with high grazing cows. The attention should also be directed on how the discovered trade-offs could be reduced, namely GWP, e.g. with feed supplements, pasture quality improvement or adapted dairy cow genetics.

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Trampling effects on leys from four seed mixtures – ground cover after grazing

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Abstract

Pasture swards can easily be damaged by trampling, causing serious problems on many farms. This study evaluated and compared swards made from four seed mixtures regarding resistance to trampling and animal selection at grazing. All mixtures contained cultivars of smooth meadow-grass (*Poa pratensis*) and red fescue (*Festuca rubra*) with/without white clover (*Trifolium repens*) as a base, plus perennial ryegrass (*Lolium perenne*) or tall fescue (*Festuca arundinacea*). The four leys were set up in a randomised block design (three replicates) and rotation-grazed for two seasons. Degree of ground cover was evaluated by spatial analysis of Unmanned Aircraft System photographs after rotations. The ley with clover, ryegrass cv. Foxtrot, smooth meadow-grass and red fescue had more (P<0.05) ground cover (76.1%) than the same mixture without clover (72.2%) or the mixture with tall fescue (72.5%).

Keywords: seed mixture, ground cover, spatial analysis, botanical composition, pasture

Introduction

Increasing herd size increases the pressure and intensity of land use and makes it difficult to maintain dense swards, especially near cattle houses. Trampling-resistant swards are crucial for grazing management, animal welfare and environmental protection. This study compared establishment rate, resistance to trampling and grazing behaviour for four seed mixtures. Ley establishment (Nilsdotter-Linde *et al.*, 2015) and grazing behaviour (Sandberg, 2015) has been reported earlier. Results from the first two grazing seasons on resistance to grazing/trampling and botanical composition are reported here.

Materials and methods

Four seed mixtures were composed to find a mixture that would give a persistent sward resistant to trampling. The mixtures were sown randomly in 12 field plots (each 12×36 m) in three replicate blocks in July 2012 (see Nilsdotter-Linde *et al.*, 2015). Two mixtures contained white clover, smooth meadow-grass and red fescue, one with perennial ryegrass (A) and the other tall fescue (B) (Table 1). Mixture C was similar to A, but without white clover. Mixture D contained similar proportions of species to C, but used amenity-type (sports turf) varieties, whereas C had forage-type varieties (Table 1). The experimental area was grazed by dry cows or heifers over a total of nine rotations, with five and four rotations and with a total of 533 and 351 grazing days (no. of animals × days on area) in 2014 and 2015, respectively.

Photos of the experimental area were taken by an Unmanned Aircraft System five times each year (before grazing started + four occasions directly after grazing rotation) to assess sward damage by calculating ground cover. One extra photo of the area was taken in June 2015 to assess sward recovery 18 days after a rotation with very severe sward damage in May the same year. The ground cover, as shown in the photos, was evaluated by spatial analysis using ArcGIS (2013). Sampling for analysis of sward botanical content was performed, on two occasions (May and August 2015) by taking small plant samples from the corners and centre of a quadrat (0.3 m × 0.3 m) thrown randomly 10 times per plot and determining dry weight of different species plot-wise. Statistical analysis of ground cover and botanical composition

Table 1. Seed mixtures used in treatments A-D.

Species		Variety (type)	Seed mixture, %			
			A	B	C	D
Trifolium repens (L.)	White clover	Undrom (small leaves)	20	20		
Poa pratensis (L.)	Smooth meadow-grass	Kupol (forage)	35	35	44	
		Julius (amenity)				44
Festuca rubra (L.)	Red fescue	Gondolin (forage)	10	10	12	
		Cezanne (amenity)				12
Lolium perenne (L.)	Perennial ryegrass	Foxtrot (late, diploid, forage)	35		44	
		Bizet 1 (late, diploid, amenity)				44
Festuca arundinacea (Schreb.)	Tall fescue	Borneo (amenity)		35		

was performed with a Mixed model with repeated measurements and autoregressive covariance structure (experimental plot 1-12) using SAS ver. 9.4 (SAS Institute Inc, Cary, NC, USA). The model for ground cover contained the variables treatment (A-D), block, occasion, the interaction treatment × occasion and a covariate (ground cover early 2014 before grazing). The model for botanical composition contained treatment, block and sampling occasion (May or August 2015).

Results and discussion

During 2014 and 2015, mean temperature May-Sept. was 14.0 and 13.4°C, respectively, and precipitation was 313 and 249 mm, respectively. Statistical analysis of ground cover showed a significant interaction (P<0.0322) between treatment (A-D) and assessment occasion, with no or minor differences between treatments on certain occasions and large differences on other occasions when heavy trampling damage had occurred. The overall treatment differences (P = 0.0527) are summarised in Table 2, which also shows the results for the occasion that showed the largest trampling damage ('worst case') during the entire evaluation period, due to heavy rainfall in late-May 2015, and recovery of the sward 18 days after this occasion. In the overall analysis, percentage ground cover was significantly higher for mixture A than B and C, while ground cover was higher for mixture D than for C. Statistical analysis of the ground cover on the worst case occasion showed that treatment B had significantly lower ground cover (46.1%) than the other mixtures (Table 2). Furthermore, after 18 days, mixtures A and D had significantly higher ground cover than mixtures B and C, but there was high (P<0.001) recovery potential in all mixtures (Table 2).

Table 2. Degree of ground cover (%) in the grazing period 2014-2015 for seed mixtures A-D. Least squares means (standard error in brackets) of ground cover (overall) and results for one separate ground cover assessment after a rotation with severe sward damage (worst case) on 5 June 2015 and recovery after 18 days of sward rest (23 June).

	Seed mixture			
	A	B	C	D
Overall				
Ground cover, %	76.1^{a} (0.81)	72.5^{bc} (0.76)	72.2^{c} (0.64)	74.6^{ab} (0.é64)
Worst case				
5 June 2015	53.3^{aA} (1.88)	46.1^{bA} (1.87)	54.5^{aA} (1.82)	55.3^{aA} (1.82)
23 June 2015	85.7^{aB} (1.88)	79.7^{bB} (1.87)	79.1^{bB} (1.82)	85.4^{aB} (1.82)

$^{a, b.}$ Values within rows with different letters are significantly different (P<0.05).

$^{A, B.}$ Values in *worst case* within columns with different letters are significantly different (P<0.001).

The analysis of the botanical composition (Table 3) revealed significantly more white clover in mixture B, with tall fescue, than in A, with perennial ryegrass (56.0 and 47.1%, respectively). This was mainly due to a significant (P<0.01) increase in clover content in mixture B, from 36.0% in May to 76% in August, while no similar increase was seen in A. This corresponds to the slower establishment and development of tall fescue compared with perennial ryegrass allowing increased white clover content in mixture B earlier reported in Nilsdotter-Linde *et al.* (2015) and in DairyNZ Farmfact (2010). The pasture selection of heifers grazing the same seed mixtures in an adjacent paddock was evaluated in a behaviour study where heifers were observed during 6 rotations during 2014 which was reported by Sandberg (2015). The results showed that the heifers spent a significantly (P<0.01) higher percentage of their total grazing time on mixture A (32.3%), compared with 26.9, 20.4 and 20.4% for leys B, C and D, respectively. Thus, the ley established from seed mixture A seems to maintain the highest ground cover (Table 2) while at the same time being a sward appreciated by grazing cattle.

Table 3. Data for two botanical sampling occasions (Occ) in May and August 2015 for seed mixtures A-D. Least squares mean and standard error (SE) for each botanical component (%).

Component	Seed mixture					Significance		
	A	B[1]	C	D	SE	Block	Seed mix	Occ
Perennial ryegrass	22.1		29.7	29.2	2.95	NS	NS	NS
Red fescue	10.8^{ab}	7.5^{b}	29.1^{cd}	25.2^{ad}	4.59	NS	*	NS
Smooth meadow-grass	12.6^{a}	14.4^{a}	19.3^{b}	21.3^{b}	1.26	NS	**	NS
White clover	47.1^{a}	56.0^{b}			1.38	NS	*	*
Herb weeds	6.2^{ab}	3.9^{a}	14.5^{b}	13.0^{b}	2.46	NS	tend.	NS
Grass weeds	0.6	0.0	7.4	1.4	2.99	NS	NS	NS
Wilted grass	0.6^{a}	0.0^{a}	0.1^{a}	9.8^{b}	2.46	NS	tend.	NS
Per. ryegrass vs tall fescue	22.1	18.2			3.69	NS	NS	NS

[1] Tall fescue was not analysed except in the comparison with ryegrass, as only mixture B contained tall fescue.

[a, b, c, d] Values within rows with different letters are significantly different (P<0.05).

Conclusions

Ground cover varied according to the weather and wear, and recovery was remarkably rapid after serious damage. The overall conclusion is that seed mixture A, with the base grasses plus perennial ryegrass and white clover, seemed to perform best with regard to resistance to trampling damage. Further studies are planned in 2016-2017 to verify this conclusion.

Acknowledgements

The Swedish Farmers' Foundation for Agricultural Research is gratefully acknowledged.

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Effects of three different pasture allocation techniques on milk yield and quality with mid-lactation dairy cows

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Abstract

We assessed milk production with 24 mid-lactation Norwegian Red dairy cows on a spring pasture dominated by timothy (*Phleum pratense*) for a period of 21 days using three pasture allocation techniques (n=8). Cows received weekly allowances at once (7 day-set-paddocking; 7SP), grazed 1/7 of 7SP allowance each day (daily-strip-grazing; 1SG), or grazed like 1SG but also had access to the previously grazed part of the paddock (daily-forward-grazing; 1FG). We hypothesized that 7SP would deteriorate sward quality and quantity over the grazing days whilst the other two treatments would provide balanced pasture quality and intake. These changes were expected to result in differences in milk yield and its components. However, changes in sward chemical composition (e.g. neutral detergent fiber, crude protein) over the grazing days in each week were not different among treatments (treatment × grazing day; $P>0.05$). Furthermore, no effect of treatments on milk yield and its components was observed. Nonetheless, the effects of grazing days over a week on milk yield and components were different among treatments (treatment × grazing day; $P<0.05$). These treatment by grazing day interaction effects, in the absence main effect of treatment, could be due to fluctuations in daily DMI among treatments over the grazing days in each week.

Keywords: grazing, pasture allocation, dairy cow

Introduction

Grazed grass with dairy cows is a cheaper source of milk production (Wright, 2005) and grazing helps dairy cows to display their natural behaviour. However, there is a requirement for supplementation with concentrates to sustain high yielding dairy cows on pasture. This comes with extra cost due to the current competing demands for cereal grains. Therefore, looking for pasture allocation techniques that could result in an optimal feed intake with needed quality to support the current level of milk production, is vital for profitable dairy farming.

Comparisons of different grazing management systems under different conditions yielded differences in DM use efficiency, milk yield in dairy cows, as well as weight gain and methane emission with steers. It was evident from the works of Bryant *et al.* (1961) that such differences could be due to changes in the attributes of the grazed diet (e.g. proportions of morphological fractions, chemical composition of these fractions and physical architecture of the grazed sward).

Here, we conducted a short duration grazing experiment with mid-lactation Norwegian Red (NRF) dairy cows to assess the effects of three different pasture allocation techniques on milk yield and its chemical composition. We hypothesized that the quality of DM consumed and the quantity to consume in a grazed horizon will deteriorate with extended grazing days in a paddock for set paddocking whereas frequent allocation of a pasture would balance DMI and its quality.

Materials and methods

Twenty-four mid-lactation (DIM±SD; 124±37) NRF cows (mean BW±SD; 572±66) were randomly allocated into six groups (two replicates of size 4) and grazed a spring pasture dominated by timothy (*Phleum pratense*) for three 7 day periods (21 days in total) in one of the three pasture allocation techniques (7 day set-paddocking, 7SP; daily strip-grazing, 1SG; and daily forward-grazing, 1FG). The first week was used as an adaptation period. Milk yield was registered daily and milk samples were taken frequently for standard chemical analysis. Grazing was augmented with a supplementation of 5 kg d^{-1} cow^{-1} commercially available concentrate feed (FORMEL Favør 90, Felleskjøpet, Oslo).

Grazed sward samples were taken to represent three time points during each measurement week (start, middle and end of grazing) and dried at 60 °C for 48 hours and milled through a 1.0 mm sieve size for standard chemical analysis. Additional samples were also taken at the beginning of the adaptation week. Dry matter availability was assessed using the raising plate meter.

Data collected over the measurement weeks were analyzed using SAS (SAS Institute Inc, Cary, NC, USA) mixed models as repeated measurement. Whenever they existed and contributed significantly to the model, day 0 measurements were used as covariates. Statistical significance was declared at $P \leq 0.05$.

Results and discussion

Data on chemical composition of grazed sward and milk yield are presented with Figure 1. Some of the measured and estimated chemical compositions were affected by grazing days in each week. As such, CP content declined in each measurement week (effect of day in a week; $P<0.0001$) regardless of the pasture allocation techniques. The decline in CP content was accompanied by an increasing NDF content in the

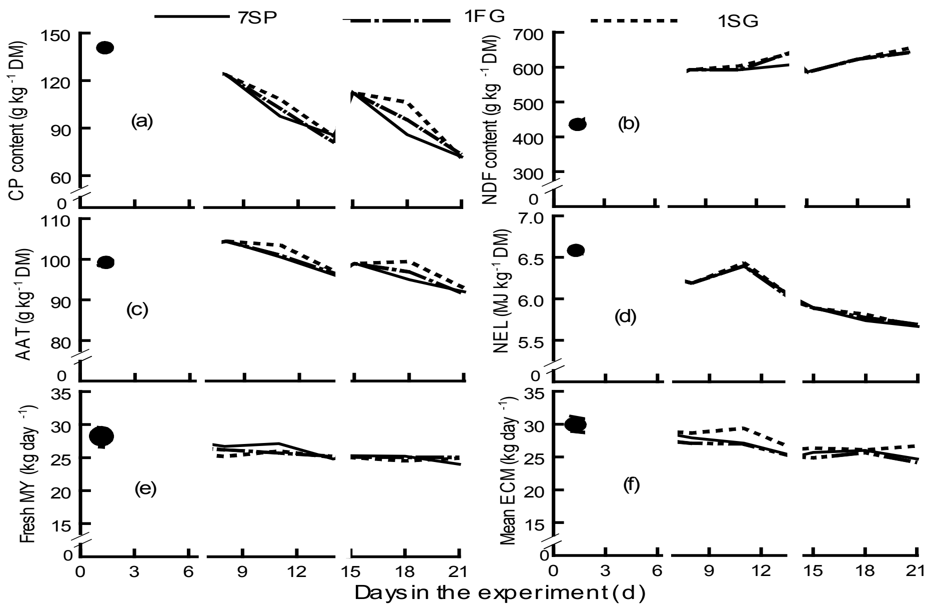

Figure 1. Chemical composition of grazed sward, [(A) crude protein (g kg^{-1} DM), (B) neutral detergent fiber (g kg^{-1} DM), (C) estimated NE_L (MJ kg^{-1} DM), (D) estimated AAT (g kg^{-1} DM)] and milk yield [(E) fresh milk yield (kg day^{-1}) and (F) energy corrected milk yield (kg day^{-1})] over the grazing period. Adaptation week value for each of the parameters is indicated with black dots on day 1. Treatments: solid, long broken and short broken lines stand for 7SP, 1FG and 1SG groups (see text for description), respectively.

grazed sward (effect of day in week; P<0.001). However, the degree to which the NDF increased was dependent on the pasture allocation technique during week 1 of the measurement week (Treatment × grazing day effect P<0.05).

The estimated net energy lactation (NEL) and metabolisable protein supply (AAT) of the grazed sward declined with grazing days in each week (P<0.0001). However, the decline was uniform for all treatment groups and there was no treatment and treatment by grazing day interaction effects. This suggested that the fast spring pasture growth and associated sward morphological developments had stronger effects on sward quality than the effects of pasture allocation techniques. This is supported by some of the observed changes in chemical composition of the grazed sward; when day 1 values are compared to day 8 and day 15 (all pre-grazing values) with drops in CP and NEL, and abrupt increment in NDF content (Figure 1).

During the experiment, fresh milk yield (25.9, 25.3 and 25.3 kg/d; pooled st. dev. 4.64) and energy corrected milk yield (mean for milk sampling dates, 26.3, 27.2 and 25.6 kg day^{-1}; pooled st. dev. = 4.49), for 7SP, 1SG and 1FG respectively, were not affected by the pasture allocation techniques (P>0.1). However, both energy corrected milk and fresh milk yields were significantly (P<0.0001) affected by day of grazing in a week. In addition, we observed interaction effects (P<0.05) between treatments and grazing days for fresh milk yield, milk lactose, protein and milk urea contents in the absence of the main effect of treatment suggestive of the fluctuations in daily DMI among treatments over the grazing days in each week.

Conclusions

In summary, the lack of anticipated differences among the treatments in sward qualities over each week, behavioural adaptations by cows to adjust DMI subject to pasture allocation techniques and probably the resilience of dairy cows to accommodate changing nutritional conditions under such short experimental periods may explain the absence of treatment effects on milk yield and its chemical composition.

Acknowledgements

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Netherlands grass monitoring network

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Abstract

To support on farm grazing management in the Netherlands a grass monitoring network was established in 2014. The aim of the network is to share and publish data on grass quality, grass growth and soil temperature in different regions of the Netherlands to serve as a benchmark. Grass quality, sward height and soil temperature were measured weekly during the grazing season of 2014 and 2015 by farmers, researchers and agricultural students. These data were used to calculate grass growth. Thus far grass growth curves of grasslands in the high output dairy farming systems of the Netherlands were not available. Publishing of the data increased the awareness of farmers on possible improvements of their grassland management, thereby supporting grazing.

Keywords: benchmark, grass growth, grazing, monitoring

Introduction

In the last decades, grazing has not been a key topic in the Netherlands. Therefore, the current grazing knowledge of farmers and advisers is relatively limited. Furthermore, farming systems in the Netherlands have changed over the years, in particular the number of animals ha^{-1} and the milk yield cow^{-1} have increased. New initiatives support farmers in their grazing management. An example is the grass monitoring network where research, education, extension, partners in the agricultural industry and farmers work together. The aim of this initiative is to benchmark grasslands by sharing and publishing data on grass quality, grass growth and soil temperature in different regions of the Netherlands.

Materials and methods

The grass monitoring network was established in 2014. Farmers, researchers and students measured sward heights and soil temperature weekly on 20-25 commercial farms spread over the Netherlands. Most farms were located in the areas with predominantly dairy farming. Sward heights were measured by rising plate meter. Average grass growth (kg dry matter (DM) ha^{-1} d^{-1}) was calculated weekly from the available measurements. Grass swards in the first week of regrowth were not used for calculating grass growth. Soil temperature (°C) was measured at 10 cm depth. In each region, one or two grass samples from grass of about 3700 kg DM ha^{-1}, were analysed weekly for crude protein content (CP, in g kg DM^{-1}) and organic matter digestibility (OMD, in %). With regression analysis a trend line with corresponding R^2 was determined for grass growth, soil temperature, CP and OMD using the polynomial function in MS-Excel. The data on grass growth, soil temperature and feed quality were published in 'De Weideman'; a weekly e-mail on grazing management for farmers and advisors.

Results and discussion

Grass growth and soil temperature are presented in Figure 1 (R^2 = 0.80 for grass growth; R^2 = 0.91 for soil temperature) and Figure 2 (R^2 = 0.79 for grass growth; R^2 = 0.94 for soil temperature) for 2014 and 2015, respectively. The growing season started about three weeks earlier in 2014 than in 2015. Maximum grass growth was nearly 100 kg DM ha^{-1} d^{-1} in both years. The 'summer dip' of 2014 occurred in July and was more severe than the 'summer dip' of 2015 that occurred in June. Grass growth dropped to 50 and 60 kg DM ha^{-1} d^{-1} in 2014 and 2015, respectively. The soil temperature followed the same pattern in 2014 and 2015, but the soil temperature of 8 °C was reached about 3 weeks later than in 2014. This corresponds with the start of the growing season in those two years. Due to ageing, grass digestibility (R^2 = 0.79) decreased during the 'summer dip' (Figure 3). Towards the end of the growing season grass was cut at a younger stage so digestibility increased. Despite the 'summer dip' the crude protein content (R^2 = 0.48) was very constant and varied between 200 and 240 g CP kg DM^{-1}.

Corral and Fenlon (1978) defined a comparative method for describing the seasonal distribution of grass production. However in this study data were collected on commercial farms. This includes the impact of e.g. a range of farm managements and especially grazing management on the data. The data were useful

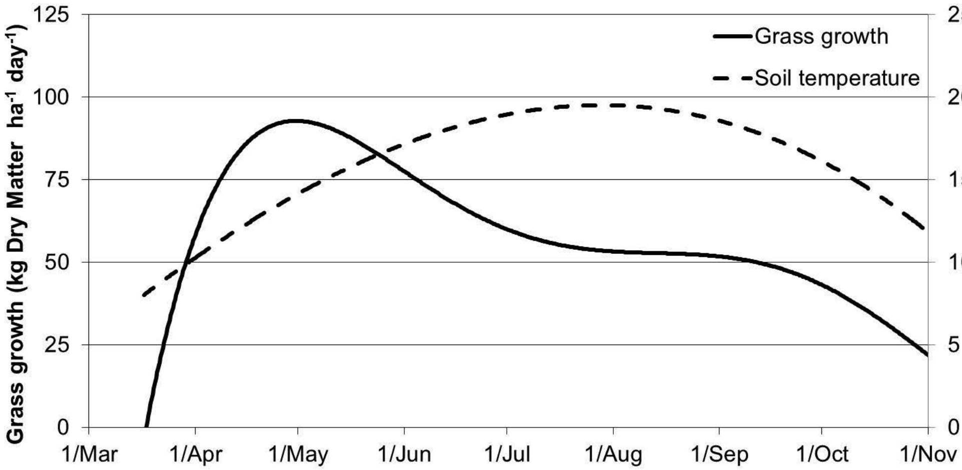

Figure 1. Average grass growth (kg DM ha^{-1} d^{-1}) and soil temperature (°C) Netherlands 2014.

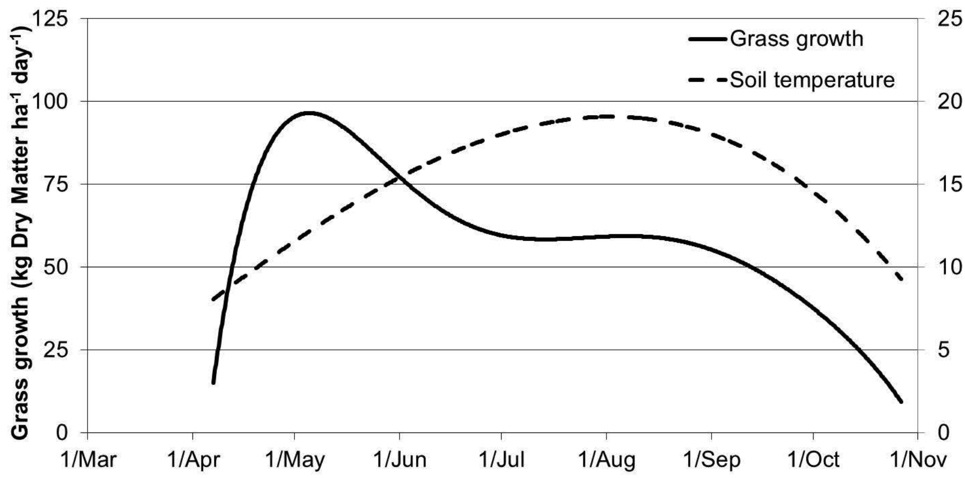

Figure 2. Average grass growth (kg DM ha^{-1} d^{-1}) and soil temperature (°C) Netherlands 2015.

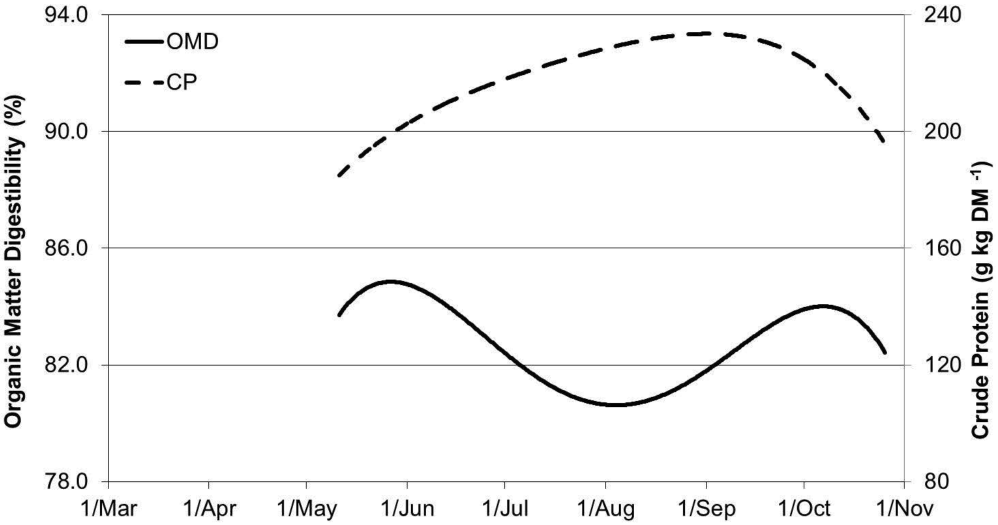

Figure 3. Average organic matter digestibility (%) and crude protein content (g kg^{-1} DM) Netherlands 2015.

in providing insight in grass growth, soil temperature and grass quality. This increased the awareness of farmers on possible improvements of their grassland management.

Conclusions

The grass monitoring network of the Netherlands provides the opportunity for farmers and advisors to benchmark grass growth, grass quality and soil temperature. These developments support grazing in the Netherlands.

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Attitudes of German grazing and non-grazing farmers towards the impact of grazing on milk production

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Abstract

Fifty four German dairy farmers answered a questionnaire concerning their attitude towards the impact of grazing on the milk production. Farmers, who provide access to pasture for their cows are not worried, that this could reduce the performance of their cows. This applies to both, grazing farms, where grass still contributes a part to the ration of the cows as well as for farms with highly restricted grazing, where the possible intake of grass is negligible. In contrast to this, farmers with all-year housing assume that grazing would reduce the milk production of cows. We also found that the performance of the cows on grazing farms is significantly lower than the performance of cows on farms with highly restricted grazing or non-grazing farms. Moreover, cows on grazing farms are generally provided with less energy from maize and concentrates than cows from non-grazing farms and farms with highly restricted grazing. We conclude that farmers with different management systems have different opinions on what they regard as a high milk production. While farmers from grazing farms are content with the milk production they achieve, farmers from non-grazing farms and from farms with highly restricted grazing pursue a higher milk production.

Keywords: dairy farmers, performance, milk yield, grazing, non-grazing

Introduction

Grazing is often associated with a reduced milk production. In order to increase their milk production, many farmers in Germany change to all-year housing and do no longer provide access to pasture for their cows. However, against this trend to all-year housing and to high milk production there are still farms where grazing contributes a substantial part of the ration. Usually, even during the grazing season the ration is not completely based on grass from grazing, but a substantial amount of energy is provided by additional feeding of maize and concentrates and grass-silage as well. Other farmers offer their cows a much restricted access to rather small pastures. These pastures are mainly used for animal welfare purposes and the actual intake of grass is negligible.

We were interested in the following questions: how do farmers from grazing farms assess the influence of intake during grazing on the milk production? Do attitudes of farmers from grazing farms and farms with highly restricted grazing differ from the attitudes of farmers with all-year housing? Does the milk production per cow differ among the three groups and if so to what extent? We followed those questions in an on-farm survey with grassland farmers in Northern Germany.

Materials and methods

In the survey three types of dairy farmers were interviewed: (1) non-grazing farms (NGF) with year round indoor housing of the dairy cows; (2) highly restricted grazing farms (RGF) where the dairy cows have a restricted access to pasture but the contribution of grazing to the total energy supply is negligible; and (3) grazing farms (GF) where grazing still contributes a substantial part of the ration, although the grazing is in most cases and to a varying degree supplemented by silage and concentrates. A farm was considered as a GF only when access to pasture exceeded three hours per day and the farm had at least 0.08 ha pasture per cow (KTBL, 2009; Van den Pol-Van Dasselaar *et al.*, 2010).

The farms are primarily situated in the German federal state of Lower Saxony (41 farms) and neighbouring districts. The farmers were interviewed during the years 2012-2014 and answered questions concerning the size of their farms, the milk production and the ration of the cows (Table 1). Additionally, they had to answer a range of questions on a 5-point Likert Scale that allowed us to infer their attitude towards the impact of grazing on milk production. These questions included the advantages and disadvantages of grazing related to, among others, milk production, supply of fodder, minerals and water. A total of 73 farmers were interviewed. Eighteen farmers did not provide sufficient or gave faulty data. Finally, a full data set from 54 farms was analysed with 18 NGF farms, 19 RGF and 17 GF.

Results and discussion

Cows of GF had a significantly lower milk production than cows of NGF and of RGF. The main reason for this might be the different rations of the cows. Cows of GF are provided with less energy from maize and concentrates than cows from NGF and RGF. Usually, cows fed with a total mixed ration produce more milk than cows which are grazing and supplied with additional concentrates (Bargo *et al.*, 2002; White *et al.*, 2002).

We can assume that the cows in all three production systems have the same genetic potential for milk production as all cows in the survey were of the Holstein-Frisian breed. This implies that cows of GF could technically yield the same performance as cows of NGF. Despite this fact farmers from GF do not think that grazing reduces the performance of their cows. They do not believe that the energy supply from grazing is too low and they do not find it difficult to provide the cows with supplemental feeds, minerals and water (Figure 1). On the other hand, farmers from NGF see grazing more critical and believe that achieving a high milk production and providing sufficient energy, water and minerals is difficult to realize in a grazing system. This is in line with findings from Denmark where farmers from non-grazing farms assumed that grazing would lead to a reduced performance (Kristensen *et al.,* 2010). Farmers from RGF have a milk production that is comparable to those of NGF but their attitude towards grazing is more positive. However, the term 'grazing' to them refers to a very restricted access to pasture which has hardly any effect on the milk production.

Conclusions

From our survey data we conclude that farmers from the different management systems have different opinions on what they regard as a high milk production. It seems that farmers from grazing farms regard

Table 1. Some data on farm structure and dairy production for the three groups of farms in the survey (n=54). Values with different superscripts are significantly different P<0.05.

Farm parameters	Production system					
	Grazing		Highly restricted grazing		Non-grazing	
	mean	**sd**	**mean**	**sd**	**mean**	**sd**
Agricultural land (ha)	117	42	230	453	245	313
Grassland (ha)	59	31	61	52	64	56
Pasture cow^{-1} (ha)	0.28^{a}	0.15	0.04^{b}	0.02	0.00^{c}	0.00
Number of milking cows	69	32	109	70	138	115
Milk cow^{-1} year^{-1} (l)	8,270^{a}	1,427	9,524^{b}	1,030	9,404^{b}	1,121
Energy from maize and concentrates cow^{-1} day^{-1} (MJ NEL)	69^{a}	34	102^{b}	20	109^{b}	23

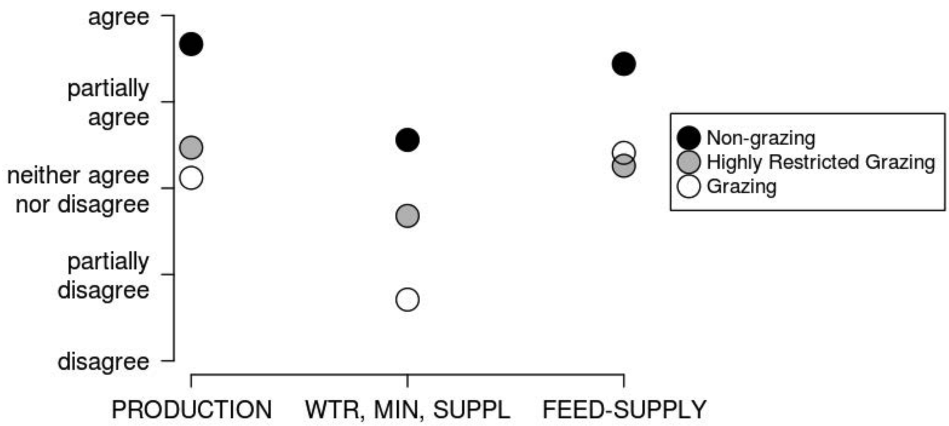

Figure 1. Attitude of farmers towards grazing: Answers on a Likert Scale from 1 to 5. Questions posed were: Grazing is challenging because (of): a reduced milk PRODUCTION due to higher energy requirements for maintenance or reduced feed intake due to adverse weather conditions; difficulties to supply cows with WTR, MIN, SUPPL (water, minerals and supplemental feeds); the FEED-SUPPLY of the pasture is not consistently high enough. Differences between GF and NGF: always $P<0.05$; difference between RGF and NGF: for PRODUCTION and FEED-SUPPLY: $P<0.05$.

their milk production as sufficient, while farmers from non-grazing farms pursue the aim of a much higher milk production and believe this to be incompatible with grazing. Farmers from farms with highly restricted grazing perceive grazing as much more positive than farmers from non-grazing farms while the milk production is comparable. When asked about 'grazing', they presumably have their very restricted system in mind which is quite different from that which farmers from grazing farms are referring to.

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The application of behaviour sensors and sward measurement to support grazing management

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Abstract

The aim of this study was to evaluate whether commercially available accelerometers for the recording of grazing time and cow activity (movements, lying, steps) combined with data on pre-grazing sward surface height, and cow characteristics (parity, days in milk, milk yield, milk composition) could be used to estimate intake of herbage by grazing dairy cows. Sixty cows which were involved in a grazing trial were equipped with two 3-dimensional accelerometers. During one test week individual herbage and total dry matter intake were measured using the n-alkane technique. The collected data were, pre-grazing sward height (cm), time spend grazing, standing and lying in seconds per quarter and number of steps per quarter. Regression analyses indicated that besides accelerometer data, inclusion of cow characteristics improved the accuracy the prediction of herbage dry matter intake. These preliminary results indicate that the use commercial available three-dimensional accelerometers may have a potential as a tool to monitor herbage intake at grazing

Keywords: cow behaviour, herbage intake, grazing management

Introduction

Three-dimensional (3D) accelerometer sensors can be used to monitor cow behaviour and differentiate between the time that cows spent grazing, ruminating, lying, walking, standing, number of movements, acceleration etc. The behavioural data derived from 3D-accelerometer sensors have shown to be very valuable as a supportive tool on dairy farms for heat detection and monitoring health problems related to claws and legs. However, it is an intriguing question whether 3D-accelerometer sensor data can also be applied to estimate herbage dry matter intake (HDMI) of individual cows at grazing. A good estimate of grazing HDMI of individual dairy cows is necessary to balance supplemental feeding (concentrates and forages) and herbage allowance (kg DM herbage cow^{-1}). Moreover, monitoring of the grazing HDMI by the herd enables assessment of herbage production and utilization. The aim of this study was to investigate if individual HDMI can be estimated from 3D-accelerometer sensor data (grazing time, standing, walking) and if the estimation of HDMI could be improved by combining grazing time with sward height measurements, and animal performance data.

Materials and methods

Sixty dairy cows (21 primiparous and 39 multiparous) were divided in 20 blocks of three dairy cows according to similarity in lactation number, days in milk, fat and protein corrected milk production and body weight. The cows of each block were randomly assigned to one of three grazing systems. Grazing systems were strip grazing (SG), 'rotational' continuous grazing system (RCG), rotational grazing on 24 1-day paddocks (DRG) which are described in detail by Holshof *et al.* (2016; this book). Cows were milked twice daily at 0600 and 1600 h using a mobile milking parlour and individual milk yield was recorded throughout the experiment. Individual milk samples were collected weekly at two consecutive milkings and analysed for fat, protein, lactose and urea (Qlip laboratories, Zutphen, the Netherlands).

All cows were individually supplemented with 4.6 kg cow^{-1} d^{-1} of a standard commercial concentrate fed in two equal portions during milking. The cows of treatment groups of cows SG and RCG were fed maize silage at rate of 6 kg DM maize silage cow^{-1}d^{-1}. Daily, per group, DM content and the fresh weight of maize silage were recorded. Daily, pre-grazing herbage allowance (PrHA; kg DM ha^{-1}) of each treatment was estimated from the sward surface height (SSH) measured by a rising-plate meter. The regression of PrHA against SSH was derived from 76 plots cut and the following relationship was obtained: PrHA (kg DM ha^{-1}) = 314×SSH (cm) – 1120; r^2 =0.93). All cows were equipped with two commercially available 3D accelerometers. One device, the 'IceQube' (IceRobotics, UK), was attached to a hind leg and recorded total standing time, lying time and number of steps. The second device, the Smarttag Neck (NEDAP, Groenlo, Netherlands) was attached to the neck and recorded grazing time in sec h^{-1} at pasture. Individual HDMI and maize silage dry matter intake (MDMI) was determined using the n-alkane method of Dove and Mayes (1991). During a 14 day dosing period, the cows received twice daily with 0.4 kg of a concentrate containing 1,046 mg kg C32 n-alkane, at each milking. During the same period the maize silage was labelled with soybean meal which was enriched with 4,165 mg C36 n-alkane, providing 161 mg C36 n-alkane kg DM^{-1} maize silage.

From day 7 to 14 of the alkane dosing period the herbage, maize silage and concentrates were sampled daily and pooled by grazing treatment for the whole sampling period. During day 7 to 14 of the dosing period, the faecal samples were, if possible, collected opportunistically from each cow twice daily after each milking. When cows were not observed defecating, then faeces were collected by rectal stimulation. The faeces samples were pooled to one sample for each cow. The concentrations of n-alkanes in grazed grass, maize silage, concentrates and faeces was analysed according to the procedures described by Abrahamse *et al.* (2008). The HDMI was calculated according to Dove and Mayes (1991) on basis of the concentrations C32 and C33-alkanes in faeces, herbage, maize silage and concentrates. The MDMI was calculated on the basis of the concentrations of C35 and C36 n-alkanes in the faeces, herbage, maize silage and concentrates.

A general linear modelling procedure (GLM) was used to predict HDMI, which was considered as the response variable (Y), with cow behaviour data (standing time, lying time, number of steps, grazing time), milk yield and milk composition, milk urea pasture data (SSH, PrHA grazing area) as explanatory variables. Stepwise multiple regression analysis was used to identify the best fitted models, based on the percentage of adjusted variance explained (adj. r^2) and best precision (lowest Mallows CP).

Results and discussion

The best fitted model for HDMI is presented in Table 1. Cow behaviour, expressed as eating time per hour at pasture, grazing area per cow, days in milk, milk yield, protein concentration and milk urea content were included in the final model for the prediction of HDMI. Standing time, lying time and number of steps did not contribute to prediction of HDMI. Inclusion of SSH did not improve the model, since the grazing area per cow and SSH were highly correlated. Inclusion of the grazing area per cow in the model was more convenient since the size of the grazed strips or paddocks already known by the farmer, and SSH required additional measurements. The proposed model resulted in a close correlation between herbage intake measured with the n-alkane technique and herbage intake predicted using behaviour sensors, sward and animal performance data (Figure 1). Inclusion of animal parameters such as days in milk and milk yield may be related to the cows' physiological status and energy requirements. The effects of milk protein content and milk urea on predicted HDMI could be related to the supply of dietary protein to the cow.

Rumen degradable protein is the main source of nitrogen for microbial protein synthesis and finally the main source of amino acids for milk protein synthesis. A surplus of rumen degradable protein is broken down to ammonia which passes through the rumen wall and is transported through the portal vein to the liver where it is converted to urea. Urea is partly recycled to the rumen with saliva and partly excreted in milk and urine. In the current experiment, grazed grass provided the largest proportion of the total rumen

Table 1. Model parameter estimates with standard error (s.e.) and t probability values of the generalized linear model for prediction of herbage dry matter intake.

Parameter	Unit	Estimate	s.e.	t (d.f.)	t pr.
Constant μ		-23.72	3.68	-6.44	<0.001
Eating time	sec h^{-1} at pasture	0.003288	0.000741	4.44	<0.001
Area per cow	ha cow^{-1} d^{-1}	-162.6	21.2	-7.68	<0.001
Days in milk		0.01433	0.00444	3.23	0.002
Milk yield	kg	0.4677	0.0438	10.67	<0.001
Milk protein	%	3.79	1.12	3.38	0.001
Milk urea	mg 100 g^{-1} milk	0.2388	0.0522	4.58	<0.001

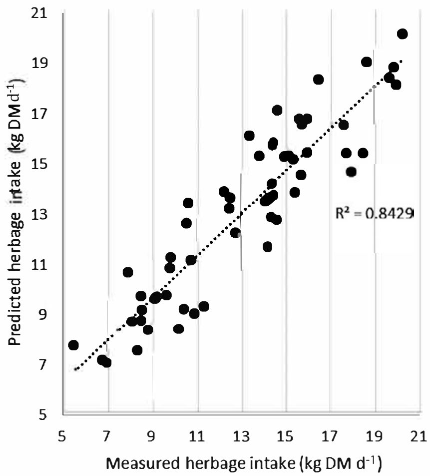

Figure 1. Correlation between measured herbage intake (n-alkanes) and predicted intake using cow and sensor data.

degradable protein intake by the cows. Therefore, the explanatory effect of milk urea content is associated with higher intake of rumen degradable protein. This also implies that the model should be used with caution when grazed grass is supplemented with feeds high in rumen degradable protein.

Conclusions

The study shows that HDMI of individual cows measured with the n-alkane can be reliably estimated from eating time, grazed area, milk yield, milk protein and milk urea. Using this information may have potential as management tools to monitor HDMI and grassland utilization. However, further research is needed to validate the proposed model with independent data.

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Effects of grazing previously abandoned grassland on performance in sheep

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Abstract

Large areas of cultivated grasslands have been abandoned in Norway and are no longer used for production. Knowing that access to spring and autumn pastures is a limiting factor for sheep farmers, this study aims at testing the effect of introducing abandoned farmland into sheep production. One sheep flock with 83(88) ewes (lambs) in 2014 and 77 (106) ewes (lambs) in 2015 was each year assigned to three treatments: (1) control; common farm procedure with a short spring grazing period before summer grazing on range pasture; (2) spring extended; a four-week extended spring grazing period on abandoned cultivated grassland before summer grazing on range pasture; (3) whole season grazing on abandoned cultivated grassland. Weight gain from spring to autumn, slaughter weight and carcass value were significantly (P<0.05) higher in lambs assigned to treatment 2, with four weeks extended spring grazing period (255 g day^{-1}, 15.7 kg, 699 NOK), compared to treatment 1 (229 g day^{-1}, 14.3 kg, 615 NOK) and treatment 3 (206 g day^{-1}, 13.2 kg, 548 NOK). The use of abandoned cultivated grassland for extended spring grazing improved weight gain and slaughter weight, while whole season grazing on abandoned grassland was the least productive option tested.

Keywords: grassland, sheep, weight gain, performance

Introduction

Common sheep husbandry practice in Norway is to keep sheep indoors during winter and free-ranged on unfenced mountain or forest pastures in summer until slaughter in autumn (Skurdal, 1997). In spring and autumn, the sheep commonly graze on fenced pastures and arable land at the expense winter feed yields. Access to grassland, particularly for spring and autumn grazing, is therefore a limiting factor in Norwegian sheep farming. Further, a general change from small to larger sheep flocks implies an increased need for access to land. Weight gain in lambs is affected by a number of different factors i.e. sex, litter size, maternal effects, as well as management and climatic factors (Dimsoski, *et al.*, 1999, Steinheim *et al.*, 2004). Most lambs are born indoors during spring and kept for a few weeks on spring pasture with the ewes before being turned out on summer range pasture. The weight of lamb when turned out on range pasture is known to affect performance (Steinheim *et al.*, 2008, Dwyer, 2009).

In Central Norway, the total farmland area was reduced by 4.9% (11,100 ha) between 2002 and 2012, of which grassland accounted for 80% of the decrease (SSB 2016). The abandoned areas will over time be encroached by shrubs and trees. This is regarded as undesirable as the quality of grasslands for sheep grazing is lost, they are important cultural landscape elements and agricultural land is scarce. Grassland is the main asset for sheep farmers and availability to grasslands is decisive for performance of production on sheep farms. In a research project, we elaborated the grass and animal production potential of abandoned grassland and the societal constraints that obstruct sheep farmers from using or getting access to such areas. The objective of this study was to test the effect of introducing abandoned grassland into sheep production.

Materials and methods

The experiment was conducted with a flock of Norwegian white spæl sheep breed with 83 (88) ewes (lambs) in 2014 and 77 (106) ewes (lambs) in 2015. The sheep farm and range pasture is situated in Tingvoll municipality in Møre and Romsdal County (N 63° 1', E 8° 8'). The sheep flock was each year assigned with respect to age of ewe and number of lambs born into three treatments: (1) Control; common farm procedure with a short spring grazing period close to the farm before summer grazing on range pasture; (2) Spring extended; a four-week extended spring grazing period on abandoned cultivated grassland before summer grazing on range pasture; (3) Whole season grazing on abandoned grassland. The abandoned cultivated grassland area selected for this study was a 15.3 ha grassland that had been unmanaged for 12 years, located in Sunndal municipality (N 62° 51', E 8° 26'). Before abandonment, the area was used as pasture for dairy cows.

All lambs were weighed at birth, at the start of spring-extended pasturing (May 23/20 in 2014/2015), after the spring-extended pasturing (June 20/19 in 2014/2015), and in autumn (Sept 14/15 in 2014/15). Lambs were 27(±6.7 std. dev.) / 25(±4.3 std. dev.) days old at start of spring-extended pasturing. Ewes and lambs in treatments 2 and 3 were transported to the abandoned cultivated grassland, a distance of 55 km, after weighing in spring. Ewes and lambs in treatment 1 were turned out on range pasture close to the farm. The animals were inspected regularly and approximately three times a week. Ewes and lambs in treatment 2 returned to the summer grazing on farm range pasture. Slaughter weight, carcass quality characteristics and carcass value were obtained from the slaughterhouse. This information was obtained from 47/77 of the lambs in 2014/2015. Ewes and lambs were monitored regularly for internal parasites. All lambs were treated with tick repellent at the beginning of the spring-extended grazing period.

Weight gain, slaughter weigh and carcass confirmation were analysed by using the MIXED procedure in SAS (SAS, 2011) to distinguish between sex, parity and age effects on weight. The initial model included fixed effects of sex, parity and age, and their interactions. There were no significant effects of the interactions, so all interactions were omitted as fixed effects from the analysis. The initial model also included year and mother by year interactions as random effects. There were no significant effects of year, and year was therefore omitted as a fixed effect from the final analysis.

Results and discussion

Weight gain, slaughter weight, carcass confirmation and carcass value were all significantly (P<0.05) higher in lambs assigned to treatment 2 compared to treatment 1 (Table 1). Weight gain, slaughter weigh and carcass value were also significantly (P<0.05) higher in lambs assigned to treatment 2 compared to treatment 3 (Table 1).

Table 1. Means of lamb weight gain spring to autumn, slaughter weight, carcass-classification of meat and fat, and carcass value of lambs with three different grazing treatments for two consecutive years of a previously abandoned grassland.[1]

Treatment	Weight gain spring to autumn (g day^{-1})	Slaughter weight (kg)	Carcass classification meat	Carcass classification fat	Carcass value (NOK lamb^{-1})
Control (1)	0.229^{a}	14.32^{a}	5.78^{a}	1.68^{ab}	616^{a}
Spring extended (2)	0.255^{b}	15.71^{b}	6.31^{a}	1.90^{a}	699^{b}
Whole season (3)	0.206^{c}	13.23^{a}	5.18^{b}	1.51^{b}	548^{a}

[1] Values followed by different letters were statistically different (P<0.05).

Extending the spring grazing period by grazing on abandoned cultivated grasslands for approximately four weeks (treatment 2) improved the performance of lambs compared to the regular farm practice of turning ewes and lambs onto range summer pasture after a short spring grazing period. Extending the spring grazing period implies that lambs are older and heavier when turned out onto summer range pasture, and the age, weight and condition of lambs at turnout on range pasture is known to affect the performance of lambs (Warren and Mysterud, 1995, Steinheim *et al.*, 2008, Dwyer, 2009). Moreover, in spring, grass growth starts earlier at lower altitudes and the extension of the spring grazing period in this study may have provided access to high quality grass for a longer period in spring, thus explaining the improved performance of lambs in treatment 1. Keeping lambs on abandoned cultivated grasslands for the whole summer grazing season (treatment 3) gave the lowest weight gain in lambs of all treatment groups. Grass quality on such land deteriorates during summer and thus poor performance is expected compared to range pasturing.

Grazing unfenced mountain and forest rangelands provides the opportunity for natural behaviour and therefore has the potential for a high level of animal welfare. It is however, also associated with factors such as disease and predators which cause animal welfare concerns and risks of high lamb losses. Providing ewes and lambs with an extended spring grazing period, and thus a farming system where older and heavier lambs are allowed on range pasture, has potential to improve performance, condition and thus animal welfare in sheep farming (Warren and Mysterud, 1995, Dwyer, 2009). Cultivated grasslands are commonly located at lower altitudes than range summer pastures. In spring, grass growth starts earlier at lower altitudes and the effect of extending the spring grazing period in this study may be explained by the access to high quality grass for a longer period in spring.

Range pastures in Møre and Romsdal are considered to be of medium quality with average lamb weight gain of 248 g day^{-1} compared to national average of 259 g day^{-1} (Norwegian Sheep Recording System, 2014). Variation in weight gain implies that there is potential for improvement and including abandoned cultivated grassland into the farming system in this study showed an increased weight gain from 229g day^{-1} to 255g day^{-1}, and an increase in the carcass value from 616 NOK to 699 NOK per lamb. One third of available rangeland grazing resources are unexploited (Rekdal, 2008). Use and access to abandoned cultivated grasslands, close to arable farm land, allows farmers to increase flock size and thereby also increase stocking rate on rangeland areas.

Conclusions

The use of abandoned cultivated grassland for an extended spring grazing period improved weight gain, slaughter weight and carcass value of lambs. Including such grassland has potential to improve animal performance and farmers' economy in Norweigan sheep farming.

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Three possible grazing systems for dairy farms with high stocking rates

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Abstract

Dairy herds in the Netherlands will increase in size the coming years, due to the abolishment of the European milk quota. As the available grazing area per dairy farm will not increase, exploring new ways of grazing with a high stocking rate has priority. At Dairy Campus – a Dutch experimental farm – three grazing systems with 6 cows per ha were tested: strip grazing (SG), one-day rotational grazing with a fixed paddock area and a fixed growing period of 23 days (DRG) and rotational continuous grazing (RCG). The daily grazing time was restricted. The grassland and animal performance of the three systems were compared. The fresh grass intake was 8.3 kg dry matter (DM) cow^{-1} day^{-1} for DRG and 6.3 kg for SG and RCG. The milk production of the three systems (25.4 kg milk) was not significantly different. All three systems are suitable for grazing with a high stocking rate, but the highest fresh grass intake was measured at the DRG system.

Keywords: grass intake, grazing, grazing system, large herds, supplementary feeding, stocking rate

Introduction

Grazing provides many economic opportunities, e.g. less contract work for making silage and for applying manure (Reijs *et al.*, 2013). Nevertheless more farmers tend to keep their dairy cows housed all year round to maintain milk production at a high level. Farmers consider it difficult to manage grazing due to highly variable weather conditions. Furthermore, they think grazing causes high losses and is time consuming. Recently the average herd size has increased, resulting in higher stocking rates on the grazing platform. These aspects, combined with milking with an automatic milking system (AMS), are causes for a decline in grazing (Van den Pol-Van Dasselaar *et al.*, 2015). To address these challenges, easy manageable grazing systems for farms with high stocking rates have been developed at the research farm 'Dairy Campus' in Leeuwarden, the Netherlands. The aim of this study was to study the grass and animal performance of three possible grazing systems with high stocking densities. In two separate experiments grass intake was measured by the alkane technique and in relation to cow movement (sensor data, Timmer *et al.*, 2016).

Materials and methods

In 2015, a grazing experiment was carried out at research farm 'Dairy Campus', Leeuwarden, the Netherlands. In this experiment three grazing systems were compared: strip grazing system (SG), a 'rotational' continuous grazing system (RCG) and a daily rotational 24 paddock grazing system (DRG). Each grazing system was executed with 20 cows, on 3.33 ha resulting in a stocking rate of 6 cows ha^{-1}. At night the cows were fed silage and concentrates. For DRG, the grass allowance depended on the grass production in 23 days on the fixed paddock. If there was enough grass, cows also had access during the night time. On the strip grazing and continuous grazing systems, cows had only access to the grass during day time between two milkings. Based on the high stocking rate it was expected that none of the three systems would produce enough grass for full time grazing during the whole season, so supplementary feed was needed. All cows received a flat rate of 5 kg concentrates per cow per day. The amount of supplementary feed as a total mixture of grass silage (30%) and maize silage (70% on dry matter (DM) base) depended on the grass growth and was fed after the evening milking. The SG system was aimed at a daily grass allowance of 8 kg DM per cow. At this allowance level it should be possible to mow for silage to obtain fresh aftermath. If the grass growth decreased, less fresh grass and more supplementary feed was provided. The RCG system had six blocks of 0.55 ha. Cows got access to a new block every day and rotated on five blocks. The sixth block was used for silage to obtain aftermath. In the DRG system, paddocks were only used for silage if the grass allowance was too high. The cows received a fresh paddock every day and the maximum rotation length was 23 days. Cows had to graze the available amount of grass in a paddock in 24 hours or less. If the grass allowance was too low for a 24 hour grazing period, cows got supplementary feed and were kept inside after the evening milking or from 22.00 hours onwards. The body condition score was measured and animal weights were recorded once every month. Differences in milk production per cow, amount of supplementary feed and gross grass production were compared between the three systems. Feed intake and milk production were statistically analysed with ANOVA. The grazing system was used as fixed factor.

Results and discussion

Fresh grass intake (FGI) (kg DM cow^{-1} day^{-1}) was calculated from the net energy (NE) requirements for milk production, maintenance and growth (NE-req, MJ cow^{-1}day^{-1}) the NE intake from supplemental feeding (NE-suppl, MJ cow^{-1} day^{-1}) and an estimated NE content of the grazed herbage (NE-grass; MJ kg DM) as FGI: (NE-req-NE-suppl)/NE-grass. The results of the three grazing systems are presented in Table 1

The DRG group had the highest fresh grass intake due to the possibility of a day and night grazing period in May. Therefore the supplementary feed intake was the lowest for this group. For the other two systems day and night grazing was not possible. The SG system needed aftermath and in the RCG system a certain grass stock was necessary to obtain enough daily grass growth. For all three systems animal condition and weight declined in the first three months of the experiment and increased after July. The herd had a calving period from December to April. There were no significant differences in condition or weight between the three grazing systems. The fat and protein corrected milk production is presented in Figure 1. The fat percentage of the DRG was slightly lower with 4.22 to 4.40% for both other groups, but not significantly different. There was no significant difference in milk production.

The rotational continuous grazing system was the most difficult to manage. There should be a balance between daily grass growth and daily grass intake, but in practice a large amount of grass was refused. It was difficult to find the optimal number of grazing hours. In this system cows were kept inside for a few weeks in July to clean up the paddocks and to get a fresh aftermath. The 24 paddock system (DRG) was easiest to manage. The grass allowance was measured every day and the grazing time was determined. The DRG system provided the highest grass intake. If the farmer is able to measure the amount of grass daily, the RCG system is easy manageable. All three systems have potential for grazing with large herds

Table 1. Grassland data and feed intake of three grazing systems for high stocking rates.

System (data from 2015)	Daily rotational	Strip grazing	Continuous grazing
Supplementary feeding (kg DM cow^{-1} day^{-1})	5.9	8.3	8.1
Daily calculated fresh grass intake (kg DM cow^{-1} d^{-1}	8.3	6.3	6.6
Number of grazing days	183	183	183
N fertilisation (kg N ha^{-1})	234	244	171
Net yield grass silage (ton DM ha^{-1} year^{-1})[a]	1.20	1.45	1.33
Total grass intake per system (ton DM ha^{-1} year^{-1})	9.2	7.0	7.3
Net yield grazing platform total (ton DM ha^{-1} year^{-1})	10.41	8.40	8.67

[a] Net grass silage is the amount of DM after field and conservation losses.

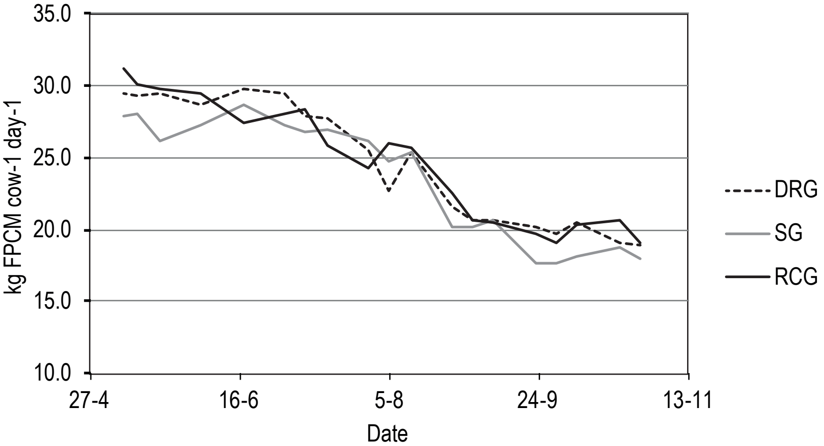

Figure 1. Fat and protein corrected milk production (FPCM) (kg cow^{-1} day^{-1}) for the grazing systems: strip grazing system (SG), a 'rotational' continues grazing system (RCG) and a daily rotational 24 paddock grazing system (DRG).

on a small grazing platform. Personal preference of the farmer will be an important reason for choosing a certain grazing system.

Conclusions

All three grazing systems, strip grazing, one-day rotational grazing with a fixed paddock area and rotational continuous grazing are suitable for grazing large herds on a relatively small grazing platform. There was no significant difference in milk production, cow weight and animal condition between the three tested systems.

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The effect of perennial ryegrass cultivars and allowance on utilisation, grazing efficiency and milk production

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Abstract

The objective of this study was to investigate the effect of four cultivars (AstonEnergy, Delphin, Glenroyal and Tyrella) on sward utilisation, milk production and grazing efficiency of lactating dairy cows at two daily herbage allowances (DHA) during the vegetative growth phase. Four cultivars were compared by offering cows two levels of DHA 15 and 20 kg dry matter (DM) cow^{-1} day^{-1} (>4cm), resulting in 8 treatments. Increasing herbage allowance from 15 to 20 kg DM cow^{-1} day^{-1} increased milk yield and solids, but reduced sward utilisation with grazing efficiency being unaffected. AstonEnergy had a longer free leaf lamina ($P<0.001$; + 4.6 cm) but also higher leaf ($P<0.001$; + 0.14) and lower stem ($P<0.01$; -0.08) than Glenroyal. This resulted in a higher sward utilisation ($P<0.001$; +12%) milk yield ($P<0.05$; +1 kg) and milk solids ($P<0.05$; +0.1 kg) than Glenroyal. Delphin was most similar to AstonEnergy in sward structure and gave similar sward utilisation and milk production, whereas Tyrella was broadly equivalent to an average of the examined cultivars and also supported an intermediate milk output. Cultivars with longer free-leaf-lamina and higher leaf content were associated with reduced number of grazing bites ($P<0.05$) and ruminating time ($P<0.001$).

Keywords: ryegrass, grazing, milk production, sward utilization

Introduction

Improved efficiency of animal production from grass is the ultimate goal of forage grass breeding for most temperate regions (Wilkins and Humphreys, 2003). Perennial ryegrass cultivars form the basis of pasture production in many temperate regions. Sward structure can vary between cultivars and has been shown to affect milk production of grazing dairy cows (Wims *et al.*, 2013), though a significant effect on milk production does not always occur (Tas *et al.*, 2006). In addition to affecting milk production, sward structure is also an important determinant of grazing efficiency (O'Donovan *et al.*, 2004) even more, where cows are managed on intensive intervals of rotational grazing which require rapid removal of herbage. Confirmation and quantification of animal responses to sward structural differences is a critical step in comparing perennial ryegrass cultivars for on farm performance. The objective of this study was to compare the milk production performance of dairy cows when grazing four perennial ryegrass cultivars varying in sward structure at two herbage allowances.

Materials and methods

Four perennial ryegrass cultivars comprising two diploids (D) and two tetraploids (T) were sown as monocultures. Cultivars included AstonEnergy (T), Delphin (T), Glenroyal (D) and Tyrella (D). Seven cows (two primiparous and five multiparous) were assigned to each cultivar during the vegetative growth phase (1 July to 28 July) at two herbage allowances: 15 and 20 kg dry matter (DM) cow^{-1} day^{-1}. This resulted in 8 treatments (4 cultivars × 2 allowances). Cows were blocked based on lactation number (3.0), milk yield (18.5 kg d^{-1}), milk solids (1.4 kg d^{-1}), protein (33.7 g kg^{-1}), fat (40.5 g kg^{-1}) and lactose (47.6 g kg^{-1}), and body weight (549 kg) and body condition score (3.0; based on a scale of 1 to 5, 1 = emaciated, 5 = extremely fat). Fresh grass was allocated to cows daily. Pre- and post-grazing sward heights were measured daily (50 heights per treatment) directly before and after grazing using a rising plate meter with a steel plate (diameter 355 mm, 3.5 kg m^{-2}; Jenquip, Fielding, New Zealand). Pre-grazing free-leaf-lamina (FLL) was measured from the highest ligula (longest leaf sheath) to the top of the leaf using a hand-ruler on 100 random tillers across each offered treatment twice weekly. Approx. 40 g of representative herbage was sampled from each treatment twice weekly to ground level; samples were then cut to the weekly post-grazing height per respective treatment and separated manually into leaf and stem fractions. Fractions were then dried for 16 h at 90 °C for DM determination. Grazing behaviour was estimated by fitting all cows simultaneously with an Institute of Grassland and Environmental Research (IGER) headset behaviour recorder after morning milking and removed prior to the morning milking 24 hours later. Individual milk yields were recorded at individual AM and PM milking. Milk solids (protein, fat and lactose) were measured twice weekly. Animal variables were analysed using PROC MIXED in SAS (2011), with grass allowance, cultivar, their interactions and pre-experiment covariate included in the model as fixed effects and animal included as a random effect.

Results and discussion

There was no cultivar × allowance interaction for any of the sward, animal or grazing efficiency variables in this study. Cows allocated 15 kg DM had a higher pseudostem proportion (P<0.05; +0.2) in the grazed sward compared to the 20 kg DM daily herbage allowances (DHA) grazed swards. Herbage allowance of 20 kg DM resulted in a lower utilisation (P<0.001; -23%), higher post-grazing height, longer post-grazing FLL and SH (P<0.001; +1.1 cm, +1.5 cm, +0.9 cm, +0.7 cm) of the grazed swards in comparison to the 15 kg DM DHA swards (Table 1). As a result, cows on the higher DHA produced significantly more milk yield and milk solids (P<0.001; +1.6, +1.1 kg day^{-1}). Although the four varieties had a similar herbage mass (kg DM above 4 cm from 12 m^2, not shown), AstonEnergy and Delphin were grazed to a lower residual height (4.1, 4.2 cm) compared to the highest, Glenroyal (+0.6 cm; P<0.001). AstonEnergy also had higher FLL (P<0.001), higher leaf (P<0.001) and lower stem (P<0.05) contents than Glenroyal. Importantly, AstonEnergy supported a higher milk yield (+1 kg cow^{-1} d^{-1}; P<0.05) and milk solids (+0.1 kg cow^{-1} d^{-1}; P<0.05) than Glenroyal. Delphin was most similar to AstonEnergy in sward structure and gave the same milk production, whereas Tyrella was broadly equivalent to an average of the examined cultivars and also supported an intermediate milk output. This is in agreement with O'Donovan and Delaby (2005) stating that cultivars producing a higher green leaf allowance impact positively on milk production. Cows grazing AstonEnergy with a longer FLL and higher leaf:stem ratio had significantly less grazing bites (P<0.001; -7083) than cows grazing Glenroyal which had increased ruminating mastications (P<0.001; +6717) compared to the other three cultivars averaging 32,329 (Figure 1). Higher stem content resulted in failure to break the barrier of standing stem despite elevated

Table 1. The effect of perennial ryegrass cultivars and herbage allowance on milk production and solids, post-grazing sward height, pre-grazing free-leaf-lamina, leaf and stem proportion and sward utilisation.

Herbage DM allowance	15 kg	20 kg	Aston[1]	Delphin	Glenroyal	Tyrella	SED	Allowance	Cultivar
Milk yield (kg cow^{-1} d^{-1})	15.1^a	16.7^b	16.3^a	16.3^a	15.3^b	15.7^{ab}	0.26	***	*
Milk solids (kg cow^{-1} d^{-1})	1.24^a	1.35^b	1.31^a	1.35^a	1.23^b	1.28^{ab}	0.03	***	*
Post-grazing SH (cm)	3.8^a	4.9^b	4.1^a	4.2^a	4.7^c	4.4^b	0.19	***	***
Pre-grazing FLL (cm)	17.5	17.5	18.7^{ab}	19.9^a	14.1^c	17.4^b	0.37	NS	***
Leaf (%)	74.0	77.0	82.0^a	78.5^{ab}	68.0^c	74.0^b	0.01	NS	***
Stem (%)	17.0	15.0	12.0^a	15.5^{ab}	19.5^b	16.0^b	0.01	NS	*
Utilisation (%)	105^a	82^b	99^a	96^a	87^c	92^b	0.01	***	***

[1] Aston = AstonEnergy.

[a–c] Means within a row with different superscripts differ (P<0.05).

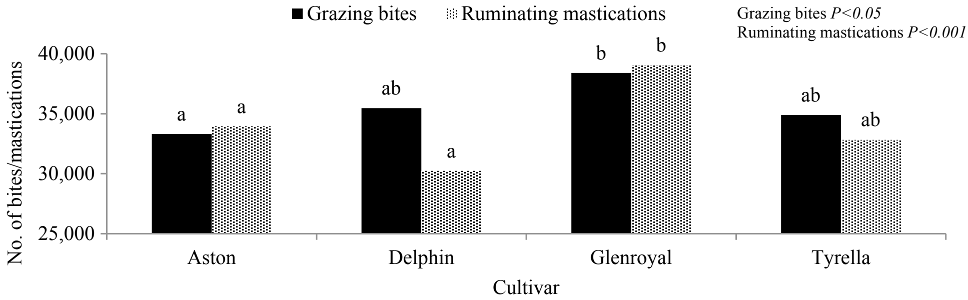

Figure1. The effect of cultivar on grazing bites and ruminating mastications of grazing dairy cows.

grazing pressure (Parga *et al.*, 2000) and impacted negatively on rumination process (Van Soest *et al.*, 1991) in turn reducing milk production.

Conclusions

To conclude, increased herbage allowance increases milk production but reduces sward utilisation significantly. Variations in sward structure in particular free leaf lamina between grass cultivars influence sward utilisation. When sward structural characteristics such as leaf : stem ratio and free leaf lamina length coupled with digestibility are differing by the magnitudes observed in this study between the four cultivars, milk production and grazing efficiency of grazing dairy cows are influenced. Improving cultivar sward characteristics such as leaf:stem ratio and free leaf lamina have the potential to improve overall efficiency of the conversion of grass to milk and improve grazing efficiency.

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Recording grazing time of dairy cows in automatic milking systems by the Lifecorder+® sensor

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Abstract

The Lifecorder+®, a uniaxial neck mounted activitymeter which is used to record grazing time of dairy cows, was validated recently (Allain *et al.*, 2014; Delagarde and Lamberton, 2015). Experiments during spring 2015 on two French experimental automatic milking systems (AMS) farms show a great individual and temporal variability. In a system with feed supplementation (Derval), daily grazing time decreases together with the decline of grass quantity and quality and the increase in the amount of supplementation. Grazing time is about 43% of the access time to pasture. In a 100% grazing system (Trévarez), where daily pasture allowance is constant, daily grazing time is about nine hours. It is longer when grass quality improves. The cows' hierarchy of access to the AMS is a predominant feeding behaviour factor. 'Dominant' animals have longer grazing time and higher animal performances (milk production, milking frequency) compared to 'dominated' animals. The use of grazing time data could be of interest in addition to other decision support tools already used by farmers (pasture probe, grazing calendars) to better combine AMS and grazing.

Keywords: grazing time, accelerometer, Lifecorder +

Introduction

The increasing number of automatic milking systems (AMS) in France and in Europe has resulted in a reduction of grazed grass in the diet of dairy cows. To improve the competitiveness of the European dairy farming by combining grazing and AMS, the European project Autograssmilk aimed to enhance the use of new technologies to succeed in this association. Recently, Allain *et al.* (2014) and Delagarde and Lamberton (2015) showed that a human activitymeter named Lifecorder+® (LC+, Kenz) could be used to assess cows grazing time. This technology was used to record daily grazing time of dairy cows in two systems combining AMS and grazing, one with supplementation and the other one in full grazing.

Materials and methods

The LC+, is a device for monitoring uniaxial acceleration. To assess grazing time on dairy cows, the sensors were mounted during two months on neck collars on cows from two AMS experimental farms in Western France (23 out of 67 cows equipped in Derval farm – Pays de Loire, 14 out of 46 cows equipped in Trévarez farm – Brittany). In Derval, the grazing system is simplified (three paddocks of 8, 10 and 10 ha) with buffer feed supplementation. In Trévarez, a rotational 100% grazing system with three paddocks per day is applied. The grazing organisations on the two farms are presented in Figure 1. The cows equipped with the LC+ were chosen to be representative of the herd in terms of production level, parity and lactation stage. The data from the sensors were converted into grazing time when the activity level was exceeding a certain threshold (Allain *et al.*, 2014). In Derval, an electronic identification system at the entrance and the exit of the barn was used to calculate the time spent by cows in the pastures. In Trévarez, the AMS was in the pastures (mobile robot), so the cows were always outside except when they were milked. Finally, 1,323 recordings were available for Derval and 518 for Trévarez. A typology of animals according to their grazing behavior was achieved through the SPAD 8.2.10® software (groups A to D in Derval and A to C in Trévarez). A principal components analysis (PCA) was used to calculate similarities between individuals. Hierarchical ascending classifications (HAC) followed by descriptions of the classes were achieved to make groups of individuals with common features. Animal characteristics

Table 1. Descriptive statistics and groups' comparison in Derval and Trévarez farms. Groups A to D in Derval and A to C in Trévarez were constituted from a principal components analysis used to calculate similarities between individuals and by a hierarchical ascending classifications followed by descriptions of the classes to make groups of individuals with common features.

Derval farm (n=1,323)						
Groups	A	B	C	D	Herd average (SD)	*P* (Fisher)
Number of animals	6	4	8	5	23	
Grazing time (min)	360^{a}	365^{a}	294^{ab}	274^{b}	320 (102)	<0.001
Number of meals	5.6^{a}	5.7^{a}	5.2^{b}	4.8^{b}	5.3 (1.8)	0.0064
Meals duration (min)	69^{a}	69^{a}	60^{b}	59^{b}	64 (23)	0.0003
Access time (min)	817 ^{a}	776 ^{a}	801 ^{a}	686 ^{b}	775 (237)	0,001
Grazing time/access time (%)	44^{a}	47^{a}	37^{b}	40^{b}	43 (14)	<0.001
Exit time	0h45^{a}	2h00^{b}	0h50^{a}	3h00^{b}	1h20	<0.001
Yield (kg cow^{-1} day^{-1})	30.0^{a}	21.2^{b}	29.4^{ab}	24.8^{ab}	27.1	0.047
Milking frequency (milk day^{-1} cow^{-1})	2.03	1.84	1.96	1.77	1.92	0.16
Trévarez farm (n=518)						

Groups	**A**	**B**	**C**	**Herd average (SD)**	***P* (Fisher)**
Number of animals	4	5	5	14	
Grazing time (min)	565^{a}	525^{b}	508^{b}	530 (79)	<0.001
Number of meals	7.2	6.9	6.3	6.8 (1.8)	0.16
Meals duration (min)	82	81	85	84 (22)	0.64
Grazing time/access time (%)	39^{a}	36^{b}	35^{b}	37 (6)	0.0004
Access time 1st paddock	8h^{a}	9h45^{b}	8h20^{a}	8h45	0.018
Yield (kg cow^{-1} day^{-1})	21.1^{a}	13.9^{b}	18.9^{a}	17.7	0.045
Milking frequency (milk day^{-1} cow^{-1})	2.01^{a}	1.57^{b}	1.77^{ab}	1.77	0.023

of the different classes of cows were tested by an ANOVA (proc GLM) with the SAS 9.4® software. The values presented (Table 1) are adjusted averages. The differences between groups were evaluated by Tuckey and Fisher tests.

Results and discussion

In a full grazing system (Trévarez), cows spent 533 minutes grazing per day in average (Table 1). In a system where they are supplemented with maize silage (Derval), the pasture access is limited and they spent only 319 minutes grazing. The difference between the two systems was also observed for the number of meals (number of grazing sequences per day – 6.8 vs 5.3) and the meals duration (grazing time per pasture visit – 84 vs 64 min). The ratio between the grazing time and the access time to pasture was quite similar in the two systems (37 vs 43%) and close from the one observed by Kaufman *et al.* (2009). In both farms, the results showed a great individual and temporal variability among the data. In Derval, grazing time decreased with the decline of grass quantity and quality and an increase in the amount of maize silage delivered. In Trévarez, where pasture allowance remains at the same level every day, grazing time is longer when grass quality is better.

Results of the PCA and the HAC are also presented in Table 1 with the comparison between the different groups of animals. In both farms, we observed that feeding behaviours enabled to bring out the place of animals in the herd social order. 'Dominant' animals, which have the first access to the AMS and therefore to the pastures, have longer grazing time and higher performances (groups A and C in Derval and group A in Trévarez) compared to 'dominated' animals (groups D in Derval and B in Trévarez). The other groups (B and D in Derval and C in Trévarez) are intermediary. Figure 1 shows the grazing kinetics of the different groups. In Derval, we can see that the 'dominant' animals have a significant meal at night and a big one when the sun is rising. The 'dominated' cows are penalized by their late access to pasture and are not able to compensate at sunrise and in the afternoon. In Trévarez, the graph in Figure 1 shows that the 'dominated' animals have a late access to the first paddock and often do not go to the 3rd one. Consequently they have to graze the left-overs of the 2nd paddock.

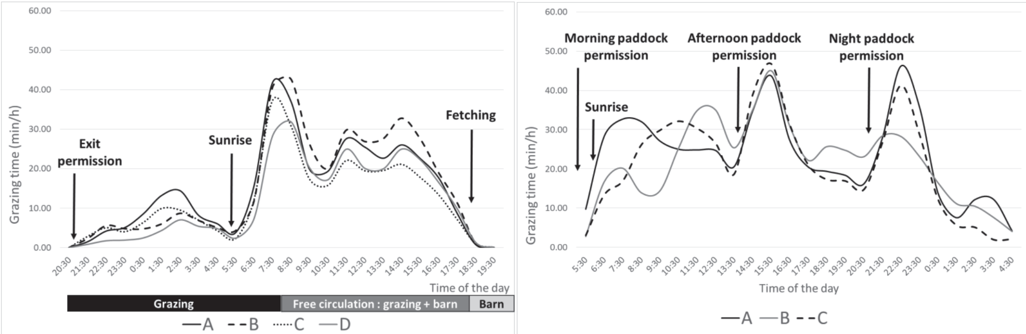

Figure 1. Grazing organisation and grazing behaviours of the cow groups in Derval (left) and Trévarez (right).

Conclusions

The use of grazing time data could be of interest in addition to other decision support tools already used by farmers to better combine AMS and grazing. In order to maximize grass ingestion, several recommendations can be provided. First of all, forage supplementation must be calibrated to the grazing allowance and not the opposite. Then, the access to AMS, therefore to pasture, could be regulated according to the animals' social hierarchy in order to give an advantage to 'dominated' cows.

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Detailed analysis of cattle behaviour on a rangeland under free range grazing system

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Abstract

Behaviour of Hungarian grey cattle was observed under range grazing conditions during four grazing seasons. The actual behaviour of cattle was recorded 3 times an hour, during investigated grazing days. Selected animals were tagged with GPS recorders which made it possible to detect their position on the pasture and to calculate the daily covered distance. Detected behavioural traits were recorded in behavioural journals (ethograms). These records allowed us to describe the animals' detailed daily routine. The daily behavioural pattern was built up from feed intake-metabolic (47.7%), social (25.9%), move (23.1%) and sexual (3.3%) behaviour traits. New traits (watch, sentinel, threefold division) were revealed and redefined (for this breed only), contrasting with previously published papers. The daily travelled distance covered by cattle, ranged between 1-5 kilometres. It was mostly influenced by the weather-front changes throughout barometric pressure dynamics. Grazing preference showed remarkable differences. Sward quality and the proximity of water had the greatest influence on the intensity of pasture use.

Keywords: cattle, behaviour, daily routine, daily travelled distance, grazing preference

Introduction

Hungarian grey cattle is a traditional breed with wide environmental tolerance that is well adapted for extensive rangelands and pastoral grazing. In the 1970's (Bodo *et al.*, 1979) the first behavioural studies revealed the general behavioural traits. These observations mainly focused on reproductional behaviour. We considered further relations from a biometerological point of view, like behavioural reactions on different weather conditions. We hypothesized that there are more behavioural traits existing than previously described. Furthermore, we hypothesized that individual weather factors have significant effects on cattle behaviour in rangeland, the grass supply has significant influence on grazing-metabolic behaviour and the spatial variation of Hungarian grey cattle is not consistent.

Materials and methods

The observed herd contained different age groups, mostly mid-age cows and their calves, but from May to August bulls made their own groups as well. We have recorded daily ethograms about the marked cows regarding behavioural traits (grazing, resting, ruminating, drinking, social interactions, moving). The herds are roaming on big pastures without restrictions. We had 10 observed cows (out of 200) marked with coloured ropes for visual identification. The cows kept 50-100 m flight-zone, therefore high vis-markers were essential. The herd did not receive supplementary feed during the grazing seasons and in winter time housing, only hay and salt was fed. The study area was 1,191 ha rangeland 5 km South-west from Hortobagy village, Hungary. Hortobagy River and two local shadoofs (dug well) provided water. The pasture had sodic soil, fertility was poor, therefore heterogenous plant cover dominated. Three yield categories (none: <100 g m^{-2}, low: 100-300 g m^{-2}, acceptable: 300 g m^{-2}<) were derived from the Hungarian grass-qualification system (Nagy, 2003).

Czako (1985) terminology has been applied to describe the animal behaviour and the behavioural traits were organised in 4 main categories. Animal behaviour was observed periodically, in every 20 minutes, and the duration of recording was approximately 5 seconds each. The most typical behaviour pattern was logged. Every grazing season 30 days (between March-November) were picked to gather behavioural and vegetation data. Meteorological data were collected from the national meteo survey database and we also made local measurements (barometric pressure, temperature). The statistical analysis was conducted by SPSS 20 software pack. Using Gere' (2003), Ungar *et al.*'(2005) and Botheras' (2010) methods, we created an activity graph, registered inactivity at the time when cows slept, rested or did not ruminate while standing. Activity was counted when feeding, drinking, movement or sexual events occured. Two type of GPS receivers have been used (Snewi Trekbox, Bluetooth, GT-750 GPS data logger). The cows grazing preference was observed at the Southern pasture (Malomhazi-pasture), during the 6 hour long (3-3 hours morning-evening) grazing period. We have counted how many cows grazed and for how long in each quadrat and assigned these in four categories (low-medium-high-very high preference). The Livestock Hour Index (LHI) (Trotter, 2009) was calculated:

$$\text{LHI (Livestock Hour Index)} = \left[\frac{Grazing\ Time\ (hour)}{Cow\ (head)}\right] \div grid/day$$

low ≤ 0,1; medium 0,1 – 0,2; high 0,2 – 0,4; very high ≥ 0,4

Results

Based on previous studies we created our own behavioural main categories (feed intake-metabolic; move; social; sexual). We have described the watch (cows, disturbed by human approach or predators, stop the actual behaviour and monitor the possible dangers) and the sentinel (chosen by dominance and position in the herd; these cows are guarding the group and continuously observing the environment) behavioural traits as the Hungarian grey cattle herd has its own organisation and hierarchy. We also specified the 3-parts dispersion within the herd. This behaviour occurs only in large fields and strongly depends from hierarchical positions. We proved that at low barometric pressure conditions (P≤1000 hPa), the herd is less active and the feed intake-metabolic behaviour is underrepresented.

There are correlations between feed intake-metabolic and move behaviours and barometric pressure (r=0.389, P≤0.05). We determined which Peczely-front types (Peczely, 1961) are the most responsible for the behavioural changes. Prior to the observation day and the following day's front were significantly affecting behaviour. The feed intake-metabolic behaviour traits appeareance follows the next day front type (r=0.445, P=0.007). The most feed intake-metabolic events happened during the anticyclone over Carpathian-basin (A). There were major effects of the anticyclones from West (Aw) (P=0.049) and North (An) as well. We have observed similar events at categories of move (P=0.004) and social (P=0.039). In both cases the high pressure local-anticyclone and the northern-anticyclone caused the highest event numbers. The average temperature, barometric pressure, humidity, wind speed and wind direction have no significant effect on the four main behavioural categories. Examining the relationship between grass supply and animal behaviour (Table 1), a significant relation was found between the event number of feed intake-metabolic group and grass supply of the area (P=0.033).

According to LHI, we determined the preferred places of residence at Malomhazi pasture. The observed animals stayed 1-2 hours at low (LHI ≤ 0.09) or moderately (LHI: 0.1-0.19) preferred cells. However, most of the visited quadrats were included in the medium preferred group. This finding can mainly be explained by the large extent of this group. The cattle spent most of their time at the northern part of Malomházi pasture (LHI ≥ 0.4), frequently camping daily here. In accordance with the literature, spatial variation was mainly influenced by proximity to water sources, spread of tussock grass and acceptable grass supply (at least 6 cm and 300 g m^{-2}).

Table 1. Relations between grass supply and the most frequent behavioural traits and groups.[1]

Grass supply category	Behavioral trait												
	Feed intake-metabolic[2]						Moving			Social			
	A	B	C	D	Average no. of events	*P*		Avg. no. of events	*P*	Watch	Scratch	Average no. of events	*P*
None	0	10	0	0	30.3±11.8	0.033*	20	15.1±2.2	0.964	0	0	30.83±2.3	0.304
Low	109	158	63	7	29.7±8.1		156			24	9		
Acceptable	255	173	121	47	52.8±5.3		226			30	31		

[1] *P* = from Kruskal-Wallis test (level of significance: $P \leq 0.05$).

[2] A = graze (standing); B = graze (in motion); C = ruminate (lying); D = ruminate (standing).

Conclusions

Incoming weather fronts affect on local pressure conditions and presumably the atmospheric electric environment as well. We proved that behaviour depends from pre-front weather conditions and Hungarian grey cattle prefer the pastures with yield 300 g m^{-2} or more. If grazing animals significantly react on pre-frontal weather systems, this could be used for management purposes as a behavioural forecast. Veterinary inspection could be less stressfull when high barometric pressure prevails.

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Comparison of grazing vs indoor feeding on environmental and economic sustainability of dairy-systems

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Abstract

DAIRYMAN was an EU-Interreg IVB project for Northwest Europe which ran from 2009-2013 involving 10 regions. A pilot farm network was set up, including 127 dairy farms covering the partner regions. Hundred and six farms practiced grazing, whereas 19 farms had only indoor feeding. The ecological and economic data of these farms, that were regularly collected from 2009-2011, were evaluated and compared in a special study. Grazing and the percentage of grass based milk production had a large influence on economic performance. Farms practicing indoor feeding had significantly lower incomes than farms practicing grazing caused by higher production costs. Size of herds, milk performance and livestock density did not affect the economic success. By contrast, environmental indicators, like e.g. N and P balances, were rated better with bigger herd size and higher milk performances. Grazing compared with indoor feeding contributed to higher farm incomes. This suggests that grass feeding and grazing systems will be more sustainable in the future given that climatic and structural conditions favour grazing.

Keywords: sustainability, milk production systems, DAIRYMAN, Northwest Europe, Interreg

Introduction

Due to the ongoing intensification processes in milk production in Europe, the socio-economic pressure caused by a high volatility of milk prices and input costs is high. Moreover, with the changing demands of the consumers and the contemporary increase in governmental restrictions concerning e.g. nutrient balances and gaseous emissions, the challenges for European milk producers are large (Langenveld *et al.*, 2007; Raison *et al.*, 2008). In this context the Interreg IVB project DAIRYMAN was established in order to strengthen milk production and to develop guidelines for successful and sustainable milk production (Aarts, 2012). However the evaluation of sustainability of dairy farms can be done with different systems, and it is necessary to collect data of farms with regard to the suitability in view of time and effort with highest possible precision. We analysed therefore data from 127 pilot farms collected from 2009-2012 in a standardized way. The objective of the present study was to answer the following questions:

- How do milk production systems differ with respect to economics and environmental parameters?
- Are there interactions between economic and environmental indicators and are they influenced by the housing systems?
- How does the change of single production factors influence the economic and environmental sustainability of the farms?

Materials and methods

This study is based on all environmental, economic and structural data gathered in the DAIRYMAN-project (Aarts 2012; Elsaesser *et al.,* 2015). The data handling and the statistical analysis were done with the programme R. The choice of indicators was made with regard to the studies of Doluschitz *et al.* (2009), Zapf *et al.* (2009b) and Elsaesser *et al.* (2015). Indicators were tested, as e.g. in the study of Foray *et al.* (2013), with a multiple regression analysis on the significance of the target attributes. The range of indicators was statistically reduced and used for the classification (Table 1). The selected indicators were compared over the years and assessed for improvement until the project end. A cross validation was used

Table 1. Indicators.[1]

Indicator	dimension
Income of milk production	€ 100 kg $FPCM^{-1}$
N-balance	N t $FPCM^{-1}$
P-balance	P t $FPCM^{-1}$
Greenhouse gas balance	kg $CO_{2\,e}$ t $FPCM^{-1}$

[1] FPCM = fat-protein corrected milk.

to check the improvement of the model by removing variables. For each indicator, means were calculated and farms were assessed to the weighted means over the three years (2009-2011). Indicator values of each farm were used to categorize farms into quantiles (0-25, 26-75, 76-100) with corresponding index values (1, 2, 3). By performing multiple linear regression analyses, it was tested for interactions between single variables. Their effects were computed to determine the marginal productivity between economic and ecological indicators.

The pilot farms were structured depending on their housing systems, the degree of diversification, herd sizes, feeding strategy and livestock density. The housing systems of the pilot farms were categorized according to the declared grazing times. Farms were considered as indoor feeding if the animals were exclusively fed in stables (18 farms). In grazing farms (106), cows had grazing times, whereas intensity and time of grazing were not investigated. For diversification of farms, 55 specialized farms with milk and forage production and 51 mixed farms with all possible combinations of earnings were selected. The average cow number of the farms was 90. 63 farms were categorized as small farms with an average of 65 cows. Large herds had 126 cows on average (59 farms). The land use intensity was classified by the number of cows per ha land used for milk production. Extensive farms (75) had a livestock density of 1.1 livestock units (LU) ha^{-1}, while intensive farms had 2.0 LU ha^{-1} in average (25 farms). Differences in feeding systems were related by the origin of feedstuffs. Criteria were the total area for silage maize, grassland and the amount of concentrates for milk production. All variables were linked with the number of cows held. A farm was called 'grass based' (95 farms) if the area for silage maize and the consumption of concentrates were below average and the grassland area was high. 25 farms were field based (opposing definition). This guarantees that grass based farms are differentiated from field based forage systems.

Results

A positive economic potential of grazing versus indoor feeding was observed, which could be explained by the higher production costs obtained of the latter farming systems (Table 2). Differentiation into herd sizes and intensity of land use, however, had no significant effects. This shows that farms with high performing cows and large herds were not able to exploit the size effects because of high costs for operational growing and production. However, the environmental sustainability indicator was positive for these farms because the production effort per kg fat-protein corrected milk is lowered in large herds, with high milk performance and intensive land use. The higher intensity allows a more efficient production which causes thinning effects and therefore an improvement of the sustainability indicator. There were significant interactions between the environmental and the economic pillars of sustainability. For example, low nitrogen balances caused higher incomes for indoor feeding and grazing systems and increased thereby the economic sustainability. Moreover, large grassland area caused significantly higher income. This led to generally positive effects of grass based milk production and mainly to positive effects of grazing as far as the climatic and structural conditions of the farms were suitable for pastures.

Table 2. Results of tested indicators.[1]

Differentiator	group	€ 100 kg FPCM^{-1}	*P*-value	kg N t FPCM^{-1}	*P*-value	GHG kg $CO_{2\,e.}$ t FPCM^{-1}	*P*-value	kg P t FPCM^{-1}	*P*-value
Housing	indoor	9.28	6.18	18.14	0.076	1,184	0.87	0.97	0.31
system	grazing	13.22	$*10^{-5}$	21.00		1,168		0.71	
Diversi-	specialised f.	14.15		16		1,121		0.52	
fication	diverse farms	10.97		25		1,214		0.99	
Farming	conventional	12.01	0.002	21.5	$8.59*10^{-9}$	1,181	0.26	0.84	0.078
system	organic	19.36		10.5		1,063		0.39	
Herd size	large	12.96	0.29	17.1	$1.15*10^{-8}$	1,112	0.075	0.60	0.04
	small	12.15		22.8		1,208		0.94	
Land use	intensive	12.91		17.5		1,129		0.13	
	extensive	12.72		22.9		1,196		1.24	
Feeding	crop based	9.61	0.0002	21.5	0.6	1,213	0.55	0.25	0.005
system	grass based	12.96		20.7		1,166		0.89	

[1] FPCM = fat-protein corrected milk.

Conclusions

There is a need to reduce the negative environmental impact and to improve the economic situation of dairy farms in the area of Europe that was sampled. The indicators show that there is a high potential for improvement in both of these factors in dairy farming. The relatively higher profitability of farms relying on grass-based grazing or biological systems and the lower N and P balances in larger farms, highlight ways in which the economic situation in dairy farms can be improved. However, the slightly higher, but not quite significant, N balance in farms relying on grazing, as compared with confined systems, requires a middle ground to be struck to balance economic and environmental considerations. Nevertheless, both the grass based feeding systems and organic production are positive in terms of income and their nitrogen balances. However, the former had higher phosphorus balances.

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Monthly, annual and altitudinal variation of forage production in a mountainous-subalpine grassland

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Abstract

In this study the results of research conducted during the years 2012, 2013, 2014 and 2015, in the mountainous-subalpine grassland 'Kostilata', an area of about 950 ha, in Theodoriana, Epirus, Greece, are presented. The grassland area was divided into three zones based on their altitude above the sea level: zone A (1,100-1,400 m), zone B (1,400-1,800 m) and zone C (1,800-2,393 m). The results showed that: (A) the quantity of forage production of the mountainous-subalpine grassland was, on average, 1,322.1 kg dry matter (DM) ha^{-1}; (B) in zones A, B and C, the forage production was, on average, 1,690.1 kg DM ha^{-1}, 1,622.1 kg DM ha^{-1} and 1,017.9 kg DM ha^{-1}, respectively; and (C) forage production was greatest in zone A during the second half of June 2014 (1,926.1 kg ha^{-1}), in zone B during the first half of July 2015 (1,817.6 kg ha^{-1}) and in zone C during the first half of August 2015 (1,250.5 kg ha^{-1}).

Keywords: mountainous-subalpine grassland, forage production

Introduction

Subalpine grasslands have a rich flora and are utilised primarily for livestock farming during summer. The quantity and quality of the forage produced is affected by climatic conditions, the physical and chemical properties of soil, botanical composition, the type of grazing animal and management (Lemaire *et al.,* 2000). Furthermore, seasonal production is affected by the distribution of precipitation, during the plant growth period (Tallowin and Jefferson, 1999). At a specific point in time, the stage of growth varies between different plant species (Bruinenberg *et al.*, 2002) and the main factors that affect the growth of plants, in natural conditions, are precipitation and air temperature (Frank and Ries, 1990). On the other hand, the change of altitude affects the prevailing climatic conditions, which further affect the properties of the soil and the botanical composition (Oztas *et al.,* 2003).

The aim of this study was to investigate the monthly, annual and altitudinal variation in forage production within three altitudinal zones of the same ecosystem in a subalpine area located in Epirus-Greece.

Materials and methods

The research was conducted in the years 2012, 2013, 2004 and 2015, in 'Kostilata' subalpine grassland. The area extends at an altitude of 1,100 to 2,393 m and it is located in Theodoriana, Epirus, in Western Greece. Sixty fixed experimental cages, 20 in each zone, with dimensions of 4×4 m, were installed, consisting of a 1 m high fence to protect plants from grazing. From each experimental cage, the aboveground biomass was collected in metal frames with dimensions of 50×50 cm placed in five different locations within the cage, according to the method of harvesting described by Odum (1971). Then, the samples were oven-dried until a steady weight and afterwards they were weighted. Forage was sampled every 15 days, when the animals were grazing. In zones A and B forage was sampled from the beginning of May to the end of August while in zone C from the middle of June to the end of August. An existing weather station (at 1,100 m) was used to collect the required climatic parameters (air temperature and precipitation) in zone

A, while for zones B and C, two weather stations were installed in May 2013, at 1,600 m and 2,050 m, respectively. The results were compared for significant differences using a one-way ANOVA test, while mean differences were checked using Tuckey's test *(P<0.05)*. Statistical analyses were performed with OriginPro 9.0 software.

Results and discussion

In zone A, the largest quantity of forage production occurred during the second half of June 2014 (1,926.1 kg ha^{-1}), in zone B during the first half of July 2015 (1,817.6 kg ha^{-1}), and in zone C during the first half of August 2015 (1,250.5 kg ha^{-1}). The maximum monthly forage production (dry matter (DM)) during the whole period of the study reached in the first half of July, on average, 1,322.1 kg ha^{-1}. In zones A, B and C the maximum monthly forage production was, on average, 1,690.1 kg ha^{-1}, 1,621.2 kg ha^{-1} and 1,017.9 kg ha^{-1}, respectively (Table 1).

The statistically significant differences observed in forage production within the same zone, could most likely be explained by the greater precipitation observed during June, July and August, in the years 2014 and 2015. Furthermore, the statistically significant differences in the quantity of forage production between each zone, as well as in the different time periods during which the forage production was greatest, could probably be explained by the different climatic conditions (precipitation, air temperature) prevailing in each zone and the variation in precipitation during the most important months of plant growth, although we did not test these relationships statistically (Table 2). The results of our study agree with those of other researchers who observed significant differences in the annual quantity of grassland

Table 1. Forage production per year, zone and dates of sampling, expressed as kg DM ha^{-1}.[1,2,3]

	Year	Dates of sampling							
		May		June		July		August	
		1st half	2nd half	1st half	2nd half	1st half	2nd half	1st half	2nd half
Zone A	2012	615.5^{a1}	$1,216.7^{a1}$	$1,424.5^{a1}$	$1,440.4^{a1}$	$1,080.4^{a13}$	726.2^{a1}	800^{a1}	932.6^{ac1}
	2013	690.9^{a1}	$1.416.9^{a1}$	$1,439.2^{a1}$	$1,502.6^{a1}$	$1,351.5^{a1}$	881.6^{a1}	549^{a1}	661.9^{a1}
	2014	609.8^{a1}	$1.544.7^{a1}$	$1,770.8^{b1}$	$1,926.1^{b1}$	$1,882.7^{b1}$	$1,606.4^{b1}$	$1,420.2^{b1}$	$1,325.5^{b13}$
	2015	570.2^{a1}	$1.434.6^{a1}$	$1,862.2^{b1}$	$1,891.4^{b1}$	$1,842.9^{b1}$	$1,500.5^{b1}$	$1,258^{b1}$	$1,069.4b^{c1}$
	mean	621.6^{a}	$1,403.2^{a}$	$1,624.2^{a}$	$1,690.1^{a}$	$1,539.4^{a}$	$1,178.7^{ab}$	$1,006.8^{a}$	997.3^{a}
Zone B	2012	59.1^{a2}	564^{a2}	885.4^{a2}	$1,325^{a1}$	$1,417.6^{a2}$	$1,166.2^{a2}$	$1,051.8a^{b1}$	$1,157.6^{a1}$
	2013	249.1^{a2}	696.9^{a2}	$1,204.2a^{b1}$	$1,409.1^{a1}$	$1,469.1^{ac2}$	$1,233.6^{a1}$	867^{a1}	995.9^{a2}
	2014	144.7^{a2}	831.7^{a2}	$1,460^{b1}$	$1,745.1^{a1}$	$1,780.7^{ac1}$	$1,519.7^{a1}$	$1,133.2^{ab1}$	900.7^{a2}
	2015	183.7^{a2}	849.8^{a2}	$1,423.2^{b2}$	$1,722.9^{a1}$	$1,817.6^{bc1}$	$1,443.2^{a1}$	$1,237.2^{b1}$	959.9^{a1}
	mean	159.1^{b}	735.6^{b}	$1,243.2^{b}$	$1,550.5^{a}$	$1,621.2^{a}$	$1,340.7^{a}$	$1,072.3^{a}$	$1,003.5^{a}$
Zone C	2012			149.5^{a3}	397^{a2}	692.1^{a3}	999.7^{a12}	978.9^{a1}	957.8^{a1}
	2013			207.9^{a2}	647.1^{a2}	871.8^{a3}	$1,078.8^{a1}$	955.9^{a1}	814.7^{a2}
	2014			169.4^{a2}	523.7^{a2}	861.8^{a2}	982.2^{a2}	$1,234.8^{a1}$	$1,157.6^{a23}$
	2015			190.1^{a3}	496.2^{a2}	797^{a2}	$1,010.9^{a2}$	$1,250.5^{a1}$	$1,105.2^{a1}$
	mean			179.2^{c}	516^{b}	805.7^{b}	$1,017.9^{b}$	$1,105^{a}$	$1,008.8^{a}$
Total mean		260.2	712.9	1,015.5	1,252.2	1,322.1	1,179.1	1,061.4	1,003.2

[1] Average rates followed by different letter (a, b, c), in the same column, in the same zone differ significantly; average rates followed by different exponent (1, 2, 3), in the same column, in the same year, but in different zones differ significantly.

[2] Means of each zone followed by different letter (a, b), in the same column, differ significantly.

[3] Results differ significantly if P<0.05.

Table 2. Precipitation and air temperature during the summer months.

G		Precipitation (mm)											
Year		2012			2013			2014			2015		
		June	July	Aug.	June	July	Aug.	June	July	Aug.	June	July	Aug.
Zone	A	34.2	24.4	124.8	58.8	36	3.6	108.6	43	49.2	76.4	34.6	99.8
	B				72	66.4	7	120.7	75.2	75.9	93.4	72	66.4
	C				65.4	63.4	4	140.4	102.4	57.4	180.4	111.8	113.6
		Air temperature (°C)											
	A	20	23	21.3	17.4	19.3	20.9	17.1	18.8	20.7	16.4	21.7	20.3
	B				14.8	17.1	18.5	14.6	16.4	18.4	14.2	18.2	18.6
	C				10.1	12.9	14.4	10.5	12.9	14.7	10.3	14.1	14.9

production, which were attributed solely to the climatic conditions that may vary from year to year (Skapetas *et al.,* 2004). Furthermore, in studies conducted in other subalpine grasslands in Greece, the maximum forage production occurred in July and August (Mountousis *et al.*, 2009).

Conclusions

In subalpine grasslands the level of forage production and the period during which the production reaches its maximum level depend on precipitation during the summer months.

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An innovative forage system to produce bioclimatic milk

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Abstract

In order to face the new challenges of dairy farming in North-Western Europe, an innovative forage system (90 ha – 72 dairy cows) has been redesigned through a collaborative and agro-ecological approach. It is based on the diversification of forage resources, with priority to their grazing. Grazed forage resources include temporary grasslands with several species and cultivars to produce biomass at different periods of the year, but also annual crops and fodder trees to address summer and winter forage shortages. Rainfed conserved forage resources are produced to cover ungrazable periods, in order to feed animals with a diet balanced in N and energy while sparing water and energy resources. To achieve these goals, annual crops such as sorghum and maize will be cropped in association with legumes, and cereal-legume mixtures are already used. Synergies between the crop and livestock systems have also been reinforced. For instance the herd reproduction is based on two calving periods centred on spring and autumn, to ensure the coherence with the availability of grazed forage resources. This new dairy system implemented in Lusignan (Vienne, France) aims to produce 'bioclimatic milk' in the context of climate change, by efficiently using natural resources and satisfying the users' expectations.

Keywords: innovative forage system, grazing, diversification, fodder tree

Introduction

Dairy farming in Western Europe has to cope with increasing societal demands (i.e. not only milk production but also environment preservation, livestock welfare, maintenance of landscape, ...) in a context of growing uncertainty. For instance, climate change will induce an increase in CO_2 air concentration and mean air temperature, but also the frequency and intensity of drought in Europe (IPPC, 2013). In areas of limited rainfall and surface water circulation, climate change will also decrease the availability and the acceptability of irrigation (Itier, 2010), amplifying the direct risk of drought on forage production. To face these new challenges in the context of dairy farming in plains areas, an innovative mixed crop-dairy system was redesigned through a collaborative approach. The participants in the design process decided that the system should aim to permit farmers to live from their milk production in a context of climate change, while saving water and fossil energy resources and contributing to a sustainable agriculture. As this new dairy system aims to produce milk in the context of climate change, by using efficiently natural resources and satisfying the users' expectations, we termed it 'bioclimatic', in analogy with the concept developed in architecture. The design method consisted to cross-fertilize the ideas of French farmer representatives, stakeholders and researchers from various disciplines, and then to render coherent their multiple proposals (Novak *et al.*, 2015). The system relies on an agro-ecological approach at the farm level aiming to optimise the interactions between forage, crop and livestock systems, so as to save water and energy resources and to secure the forage production. This is a new approach since so far, the principles of agroecology have not been considered in the dominant dairy farming systems of Europe (Dumont *et al.*, 2012).

The forage system (90 ha) was entirely redesigned to produce the fodder necessary to feed a dairy cattle herd (72 milking cows, and replacement heifers) without irrigation and with limited use of mineral nitrogen fertiliser, whatever the climatic hazards. The dairy system has been implemented since 2013 on an INRA facility located in Lusignan (Vienne, France), south of the French leading dairy regions, in an area already affected by summer droughts.

This paper presents the innovative forage system designed for this agro-ecological dairy system called OasYs, and gives an overview of its livestock system.

An innovative forage system giving priority to grazing

The forage system is based on the diversification of forage resources, with priority given to their grazing. One of the many advantages of grazing is to be a method of feeding particularly energy and water saving, grazed forage being generally balanced in N and energy, rich in water content, and animals doing themselves harvest and fertiliser application. Enhancing diversity in farming systems is also an effective way to promote critical ecosystem services such as nutrient and water recycling and pest and disease control, while strengthening the resilience of the whole system (Kremen *et al.*, 2012). The forage system (90 ha) is based on three crop rotations, two of them providing grazed resources and the third one, conserved forage resources. Grazed rotations have the objective to allow grazed forage resources to meet 100% of the herd requirement in spring, 50% in summer and autumn and 25% in winter. They consist of five years of multispecies grassland and two years of grazable crops. All the terms of these two rotations are present simultaneously on 14 fields, with each receiving the rotation with a one-year lag. When the two rotations will be fully established, dairy cattle will have the opportunity to graze ten grasslands of diverse compositions and ages. This diversity in species and cultivars between grasslands has the objective of providing grazable forage resources under a wide range of climatic conditions and all the year round. So, instead of sowing the 'ideal' grassland adapted to specific climatic conditions, we decided to set up a diversity of multispecies grasslands that the farmer will assemble, so as to make up a fodder menu adapted to the climatic condition of each season.

These two grazed rotations differ mainly by the objectives assigned to the two years of annual crops. In the totally grazed rotation, they have to address the shortages of grass generally observed in summer (e.g. by use of millet-clover) and winter (e.g. by use of fodder beet) so as to graze all the year round. In the other grazed rotation, whose fields are located farther away from the milking parlour, these two years have to secure the forage autonomy by providing dual purpose crops (sorghum and cereal-legume mixtures), which can either be grazed or harvested, depending on the livestock needs and climatic hazards. Trees and shrubs have been planted on these two rotations (Novak *et al.*, 2016) to provide fodder in periods of low grassland production (summer and autumn) but also to shelter cattle and to improve the efficient use of natural resources.

The third rotation (4 years of multispecies meadow followed by maize, wheat, sorghum and protein-rich plants) provides forage stores, grains and straw for the livestock system, and is not grazed, because it is too far away from the milking parlour. As for the grazed rotations, all the years of the rotation are present simultaneously. To feed animals with a diet balanced in N and energy with limited use of mineral N fertiliser, forage grasses are cropped in association with legumes, either in meadows or in the annual forage crops. Crops adapted to drought are used, such as sorghum or lucerne. The diversification of management practices and cutting frequency, adapted to climatic conditions, allows the provision each year of a diversity of stores in terms of quality and type (hay, wrapping, silage), and to secure the forage system.

This forage system has been implemented from September 2013, and the first results show an increase in the use of grazed resources (Table 1).

A consistent livestock system

The herd reproduction is based on two calving periods centred on spring and autumn, to ensure the coherence with the availability of grazed forage resources. The lactation length is extended to 16 months, in order to limit the non-productive time during cow lifetime and the associated negative environmental

Table 1. Some grazing characteristics of the OasYs system experiment.

	2013	2014	2015
Extent of the grazing season	4 March – 5 Dec.	5 April – 3 Dec.	18 Feb – 31 Dec.
Total number of grazing days (full-time grazing days)	269 (81)	259 (132)	286 (134)
Total grazed area (ha)	29.4	34.7	43.7

impacts. As this dairy system needs more robust cows, with good reproduction capacity, and well adapted to grazing and to forage resources of contrasting quality, a three-way crossing of dairy breeds (Holstein, Scandinavian Red, Jersey) has been set up since June 2013 on the previous Holstein herd.

Perspective

This new dairy system is being tested since June 2013 at the farm scale for a period of about twenty years. This reality check will permit to verify the coherence of the whole system and to test its robustness, and if necessary, to adjust some of its elements. The agronomic and zootechnical performances as well as the environmental, economic and social dimensions will also be evaluated at a pluriannual scale, to test if the increase of diversity allows conciliation of good production levels and high environmental performance, and to improve the resilience of the whole system. As the experimental site is localised south of the current leading dairy regions of France, it will allow key elements in these dairy systems to be adapted to mid-term climate changes. This new system gives also food for thought to design agro-ecological forage systems for dairy farming in North-Western Europe. Scientists are invited to use this experimental system to broaden their knowledge on low-level input farming systems. This project also aims to give farmers the keys for a successful transition, as well as operational results on the new practices tested in the forage system.

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Role of pasture-based diet in modulating some meat nutritional traits of young Sarda bulls

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Abstract

To highlight the effects of pasture and hay and concentrate-based diets on α-tocopherol content and fatty acids composition of intramuscular fat of young Sarda bulls at equal growth rate, a study was undertaken. Twenty-eight animals, homogeneous for live weight (288.7±29.0 kg, mean±s.d.), body condition score (2.69±0.18) and age (355±25 days) were divided into 4 groups: one group (PAS) was fed 24 h day^{-1} at pasture without supplementation; three groups were confined and fed natural pasture hay *ad libitum* and an increasing daily dose of concentrates (2.5 kg head^{-1} HC1, 3.5 kg head^{-1} HC2, and *ad libitum* HC3). This paper, part of larger one, only reports the results for group PAS and HC2, which showed an undifferentiated growth rate (PAS 0.75±0.05 and HC2 0.78±0.05 kg head^{-1} day^{-1}). The pasture-based diet increased (P<0.05) the content (mg g^{-1} of total lipids) of linolenic acid (22.3±0.8 vs 10.9±0.8, Lsmeans±SE), ω3 (31.9±1.2 vs 19.7±1.2), α-tocopherol (3.9±0.1 vs 2.1±0.1 mg kg^{-1} of meat), and lowered n-6/n-3 ratio (3.6±0.2 vs 5.9±0.2). The results confirm that pasture can result in a better meat quality with putative benefits for consumers' health.

Keywords: grazing, cattle, fatty acids, α-tocopherol, local breeds.

Introduction

Due to the high cost of fattening based on concentrates, Sardinian farmers prefer to sell calves at weaning to the fattening centres in the North Italy and this explains the low self-supply rate of beef in Sardinia, which hardly reaches 48%. The use of pasture in the fattening of calves could decrease the feeding costs and improve the meat quality for human health (Daley *et al.*, 2010; Realini *et al.*, 2004). However, in many of these experiments dietary effects are confounded with differences in carcass weight at slaughter or with animal age (French *et al.*, 2000). This happens when animal fed high-energy diets, usually based on grains, are heavier, fatter and younger at slaughter than counterparts fed forage-based diets (Steen *et al.*, 2003). In order to isolate the effects of diet from those of nutrition level on some meat traits of young Sarda bulls, a study was carried out, comparing animals with similar growth rates.

Materials and methods

The study was carried out during 2010 (May-July) at the experimental farm of the Agricultural Research Agency of Sardinia (AGRIS Sardegna) located in Foresta Burgos (latitude 40°25'N, longitude 8°55'E, altitude 850 m a.s.l.). The site is characterized by Mediterranean climate, with minimum and maximum mean temperatures of 1.7 °C (January) and 28.0 °C (July), respectively and average annual rainfall of 905 mm, mostly from November to March. A 7 ha natural pasture based on oak tree forest, manly *Quercus pubescens* L was grazed by 28 young Sarda bulls, divided into 4 groups (7 heads per group) homogeneous for live weight (LW, 288.7±29.0 kg, mean ± s.d.), Body Condition Score (BCS, 2.69±0.18 five-points scale, Lowman *et al.*, 1976) and age (355±25 days). One group was fed 24 h on pasture (group PAS), without supplementation. The other three groups (HC groups) were kept in roofed pens and fed daily with natural pasture hay (*ad libitum*) and commercial concentrate at 3 different levels (kg head^{-1}): 2.5 (group HC1), 3.5 (group HC2) and *ad libitum* (group HC3), offered twice a day. The animals were slaughtered at fixed age (416±25 days). 24 h post-mortem, samples of *Longissimus dorsi* were removed between sixth and seventh thoracic vertebrae from each left half-carcass. The α-tocoferol (Vitamin E) content and the fatty acids composition were determined in intra-muscular fat (Addis *et al.,* 2013). The results reported hereunder refer only to the groups that showed similar growth rates during the experiment (PAS and HC2). These data were analyzed with lme procedure of R using a linear mixed-effects model with diet as fixed effect and animal as random effect.

Results and discussion

PAS and HC2 bulls showed similar LW (PAS 340±9 and HC2 336±9 kg LW, P=0.74) and growth rates (PAS 0.75±0.05 and HC2 0.78±0.05 kg head^{-1} day^{-1} P=0.67).

The PAS group showed the higher value of the α-tocopherol content (Table 1), falling in the optimal range to extend the shelf life of retail beef (0.30-0.35 mg 100 g^{-1} of fresh meat, Geay *et al.*, 2001). The α-tocopherol plays also a role in preventing coronary heart diseases, hampering the synthesis of nitrosamines (Daley *et al.*, 2010). While intramuscular fat from HC2 was higher in oleic acid; α-linolenic and eicosapentaenoic (EPA) acids were higher in pastured-animals, likely because of the fatty acid composition of the diet. The α-linolenic acid is the major fatty acid in grass lipids (Scollan *et al.*, 2006) and EPA arises from its elongation and desaturation. Both contribute to increase the n-3content, in meat from PAS group, lowering the n-6/n-3 ratio that was below the threshold (n-6/n-3<4) indicated by the Committee on Medical Aspect of Food Policy to prevent some cardiovascular diseases. Unexpectedly, the CLA 9c 11t content was lower in PAS group, mirroring the trend of its precursor, vaccenic acid, which

Table 1. The effect of diet on individual fatty acid proportions in intra-muscular fat of *Longissimus dorsi* at 6th/7th rib and α-tocopherol content of young Sarda bulls. (Lsmeans±SE).[1]

Fatty acid (mg g^{-1} total lipid)	HC 2	PAS	SE	*P*-value[2]
C16:0	128.48	119.06	5.88	ns
C18:0	137.32	133.64	6.85	ns
C18:1 11t (TVA)	17.38	12.95	1.46	0.05
C18:1 9c oleic acid	162.04	133.88	7.84	0.05
C18:2 9c,12c (linoleic acid)	97.50	97.26	5.49	ns
C18:3 9c,12c,15c (linolenic acid)	10.94	22.35	0.81	0.001
CLA 9c,11t	3.02	1.99	0.21	0.01
C20:4 5c,8c,11c,14c	18.58	18.88	1.60	ns
C20:5 5c,8c,11c,14c,17c EPA	0.26	0.49	0.03	0.001
C22:5 7c,10c,13c,16c,19c DPA	7.96	8.60	0.55	ns
C22:6 4c,7c,10c,13c,16c,19c DHA	0.52	0.54	0.06	ns
SFA	277.16	262.57	13.02	ns
MUFA	179.43	146.83	8.85	0.01
PUFA	139.02	150.33	7.59	ns
UFA	318.45	297.16	4.58	0.01
Σn-3	19.68	31.97	1.25	0.001
Σn-6	116.08	116.14	6.91	ns
Σn-6/Σn-3 ratio	5.92	3.63	0.21	0.001
PUFA/SFA	0.51	0.60	0.06	ns
α-tocoferol (mg kg^{-1} of meat)	2.11	3.94	0.12	0.001

[1] SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; UFA = unsaturated fatty acids.

[2] ns = P>0.05.

is one of the major intermediates formed during rumen biohydrogenation of C18 polyunsaturated fatty acids (PUFA) (Alfaia *et al.*, 2006). We cannot exclude that PAS diet included species containing plant secondary metabolites (PSM) able to curb PUFA lipolysis and biohydrogenation in the rumen (Cabiddu *et al.*, 2009). Flowers or other species such as some daisy plant can contain other PSM such as terpenes, or complex enzymes such as polyphenol oxidase which can modulate fatty acid metabolism in the rumen (Buccioni *et al.*, 2012).

Conclusions

At similar nutritional level and equal finishing age, the grazing bulls, thanks to the characteristics of the ingested pasture, produced meat of better quality for consumers' health as compared with that obtained under feedlot conditions. This could result in a greater appreciation of grass-sourced meat by the consumers.

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Success of *Brachypodium pinnatum* in oligotrophic grasslands: seasonal allocation patterns of nutrients and metals

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Abstract

Grassland systems have evolved under the effect of fire and herbivory. Disruption of a sustainable disturbance regime poses a serious threat to the maintenance of diversity and may lead to degradative processes. In Western Pyrenees, the native perennial tall-grass *Brachypodium pinnatum* is experiencing an increased expansion that is accompanied by a severe loss of sympatric species. As a consequence, low-diverse and stable covers develop which are very productive despite the prevailing stressful conditions (high-altitude, extremely acidic and poor soils, metals solubilised). In order to gain insight into the mechanisms explaining the temporal stability of degraded covers, we selected eight populations of the species developing within the same regional area, four in grazed-diverse and four in ungrazed-degraded covers, and analysed the aerial and subterranean dynamics of mineral and metal allocation during a growing season. The belowground reservoir played a key role in the translocation and reallocation of nutrients and in the accumulation of toxic metals (mainly aluminium), which made the species very competitive. The results are discussed focusing on the role of herbivory and on the build-up of the vigorous, complex belowground system.

Keywords: highlands, *Brachypodium pinnatum*, oligotrophic grasslands, nutrients, metals

Introduction

Open highland landscapes in Europe are experiencing deep changes linked to the global change and, particularly, to the rural exodus and land-use abandonment. In Pyrenees, shrub encroachment and afforestation are frequent processes occurring all over the range, due to the relaxation of grazing practices (low stocking rates and lack of guided grazing). As a consequence, the use of traditional tools to control grazers' rejections, such as controlled burnings, is nowadays increasing in both intensity and frequency.

The expansion of the native tall-grass *Brachypodium pinnatum* in Western Pyrenees is likely related to the increased fire regime and decreased herbivory (Canals *et al.*, 2015). The species grows usually in diverse chalk grasslands but conditions of change make it very competitive and aggressive to sympatric species (Bobbink and Willems, 1987; Hurst and John, 1999). The success of *B. pinnatum* gives rise to extensive areas with an herbaceous cover fully dominated by the species, which are stable and lack of successional processes towards shrub or tree encroachment, despite the absence of herbivores. This research was planned to understand the mechanisms explaining the persistence of these covers, which create a situation very difficult to revert. We analysed the performance of eight on-site populations of *B. pinnatum* growing in two contrasting covers, as a sympatric component of rich, grazed grasslands or as a dominant component of low-diverse, ungrazed covers. Both community types were similar in terms of abiotic factors – climate, substrate and soil – but differed in their recent historical management. We studied the performance of the populations in terms of biomass production and analysed nutrients – nitrogen (N), phosphorus (P) and potassium (K) – and metal – aluminium (Al) and lead (Pb) – dynamics and compartmentalisation along a growing season.

Materials and methods

The research was done in Aezkoa valley commons (42°57'N 1°10'W and 43°3'N 1°13'W; 800-1,450 m a.s.l.). The area, included in the SIC Roncesvalles-Selva de Irati (ES0000126), encompasses a mosaic of beech forests, heathlands and grasslands and is dominated by a cold, rainy and misty climate (Tm= 9.3 °C; *P*=1,856 mm per year). Rangelands support nowadays a free mixed grazing 6 months per year with low stocking rates (<1.2 AU ha^{-1}). Relaxed grazing has promoted in the last decades an uncontrolled use of fire in the most remote areas.

In spring 2013, we selected eight sites that encompassed the variety of *B. pinnatum* covers in the area: four low-diverse, *B. pinnatum* dominant covers (80-100%) and four high-diverse communities (30-50% cover of *B. pinnatum*). Despite the contrasting grassland communities, soils were similar between covers for the main physical and chemical traits. Soils were very organic (9% OM) and acidic (pH in water=4.74) and developed on sandstones and calcareous clays. At each site, a sampling itinerary of 600-800 m was scheduled, in which 5 clumps dominated by *B. pinnatum* vegetation were selected. At each point, a 15×15×15 cm block of soil and vegetation was collected and kept cool. In the laboratory, aerial (necromass and green) and subterranean tissues (including roots and rhizomes) were separated from the soil. Tissues were oven dried, finely sieved and sent to the *CEBAS-CSIC* for main nutrients analyses. N was determined by elemental combustion and the rest of elements by ICP-OES atomic absorption spectrophotometry (ICAP 6500 model, Thermo Electron IRIS Intrepid II XDL Duo). Field samplings and laboratorial procedures were repeated three times during the season, after snowmelt (late April), in midsummer (early July) and in autumn (late September). As a whole, 120 blocks were processed, 40 blocks per sampling date.

Statistical analyses used R software (RC Team, 2011). The models included cover type and sampling date as fixed factors and sampling point nested within site as random factor. A correlation structure to account for the repeated measures on the same sampling plot and/or a variance structure if the residual spread differed between sampling dates were included if necessary. Differences among LSM of sampling dates and comparisons between vegetation cover types within each sampling date were done by Tukey tests (lsmean package).

Results and discussion

Results show *B. pinnatum* as a high-productive grass, with a considerable belowground system formed by a dense network of rhizomes and roots which is most of its total biomass. Herbivory appears to play a key role in the above and belowground biomass dynamics. Early in season, stands in low-diverse, ungrazed covers averaged 400 g m^{-2} of necromass compared to 100 g m^{-2} in high-diverse, grazed covers. The belowground biomass was much higher in stands of low-diverse covers (c. 850 g m^{-2}) than in the rest of sites (c. 500 g m^{-2}). Because of the absence of defoliation by grazing, low-diverse covers accumulated high amounts of aboveground tissues, which appeared to influence belowground patterns in the same way.

Nutrient concentrations in aboveground tissues varied temporally (P_{date}<0.001 for all nutrients) and showed similar seasonal patterns, reaching a peak in spring and decreasing along the season (Table 1). Nutrients in belowground tissues also varied temporally (P_{date}<0.001 for N and K, P_{date}<0.008 for P) showing the highest concentrations in spring. The results suggest that herbivory influenced the timing of internal nutrient regulation, and particularly the remobilization of nutrients to the rhizome, since different patterns of temporal translocation were found between covers. Aboveground concentrations of N were higher in high-diverse compared to low-diverse covers most of the time (P_{cover}=0.004, $P_{interaction}$=0.051), whereas the concentrations of P and K slightly differed between covers, except at the end of the season ($P_{interaction}$=0.006 for P; $P_{interaction}$<0.001 for K). As a consequence, at the end of the growing season, individuals in high-diverse, grazed covers have more nutrient-enriched aboveground tissues than individuals in low-diverse, ungrazed covers. Patterns of metal allocation in *B. pinnatum* were consistent for the two elements studied (Al and Pb) and differed radically from patterns of nutrient allocation. Belowground structures accumulated the highest quantity of metals (Table 1), that were translocated to aerial parts in small amounts along the season, (P_{date}>0.001 for Al, P_{date}>0.001 for Pb).

Table 1. Nutrients (N, P, K) and metals (Al, Pb) concentrations in aboveground and belowground tissues of plants of *B. pinnatum* growing in low-diverse and high-diverse covers. In spring samplings, necromass (previous season material) was separated from green tissues (early season material). Values are the mean ± standard errors for 20 replicates.

	Cover	previous	Aboveground			Belowground		
			early	mid	late	early	mid	late
N (%)	low	0.9±0.04	2.68±0.05	2.02±0.06	1.39±0.06	1.01±0.04	0.82±0.03	0.94±0.04
	high	1.10±0.04	2.82±0.09	2.28±0.05	1.75±0.06	1.11±0.04	0.80±0.03	0.86±0.03
P (%)	low	0.03±0.002	0.16±0.01	0.12±0.01	0.07±0.003	0.05±0.003	0.05±0.002	0.06±0.003
	high	0.04±0.001	0.16±0.01	0.14±0.01	0.10±0.01	0.07±0.01	0.05±0.01	0.06±0.01
K (%)	low	0.17±0.01	1.86±0.05	1.60±0.11	0.84±0.04	0.17±0.01	1.86±0.05	1.60±0.11
	high	0.25±0.02	1.67±0.06	1.53±0.08	1.07±0.06	0.67±0.04	0.52±0.05	0.41±0.03
Al	low	1,696.61±223.37	329.85±42.58	979.66±157.27	1,198.93±306.32	2,958.75±233.54	3,558.48±726.76	2,704.55±208.19
(mg kg^{-1})	high	1,876.22±372.69	394.01±45.62	1,524.70±233.49	1,867.55±411.63	4,283.71±579.53	2,350.17±186.10	2,850.45±231.22
Pb	low	2.99±0.22	0.61±0.07	0.91±0.13	2.08±0.24	4.52±0.38	4.14±0.65	5.07±0.74
(mg kg^{-1})	high	2.94±0.30	0.56±0.05	1.22±0.19	2.62±0.34	4.94±0.84	2.83±0.30	3.93±0.32

Conclusions

Altogether, the results suggest that belowground organs perform complex functions, not only as foraging structures and nutrient reservoirs but also as efficient sinks for the harmful metals that may solubilize in acid soils.

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Materials to prevent trampling damage on pasture areas subjected to high dairy cow traffic

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Abstract

Trampled and muddy surfaces at paddock entrances can create problems with cow traffic and milk quality; therefore causing problems for many dairy farmers. The aim was to assess the benefits of two ground stabilizing materials, bark chips and armoured mats, when compared to taking no action. The two-year field experiment used a randomized complete block design with three replications of each treatment plot (6×6 m) at the entrance to nine paddocks. Assessment of trampling damage was performed on 5-6 occasions per season. The number and depth of holes were documented and used to calculate a surface index with which to score degree of damage. The number of cow passes and weather were documented continuously. There was no significant effect of the number of cow passes (2,000-7,000) on the surface index. The armoured mats had a significantly lower surface index indicated less damage due to trampling than bark chips. However, the armoured mats had the highest cost and economic benefit is uncertain as their durability is presently unknown. Taking no action resulted in a significantly lower surface index than compared to bark chips, and was deemed a sufficient measure for current dryer weather conditions than normal and rotational grazing system.

Keywords: cow traffic, pasture, trampling, material, walkway

Introduction

A common problem on dairy farms during the grazing period is that surfaces in pasture walkways, entrances to paddocks and around water troughs become trampled and muddy. Problems associated with these damaged surfaces can, for example, cause problems with milk quality and result in non-functioning cow traffic (Lindgren and Benfalk, 2004; Bergsten, 1997). Ground stabilizing materials can be used to avoid problems like these by stabilizing high loaded sub-surfaces and protecting the ground from damage. The aim of this project was to evaluate and compare the function and cost of two stabilizing materials, bark chips and armoured mats, with each other and with taking no action.

Materials and methods

The field experiment was included in an existing rotational grazing system for a dairy herd of approximately 280 heads at the Swedish livestock research centre east of Uppsala. The field experiment (2013-2014) used a randomized complete block design with three replications of each treatment plot (6×6 m). The three treatments were applied at the entrance to grazing paddocks used by dairy cows, a sub-area that can be representative of high load trampling. The control treatment consisted of a paddock entrance that had not been subjected to any stabilizing measures. The bark chips were placed in a 25-30 cm deep layer on a sheet of geotextile. The armoured mats with geotextile glued underneath were placed on the ground and fixed with a layer of crushed limestone. The surfaces were then evaluated with an index calculated from the number and size of holes in the surface of the treatment plot (Lindgren and Benfalk, 2004; Lindgren and Lindahl, 2007). The number of holes in the depth categories 0.02-0.05 m, 0.05-0.1 m and >0.1 m were counted in 30 randomly chosen squares of 0.5×0.5 m, representing 25% of the treatment plot. The number of holes in each category was then multiplied by a factor, i.e. 1: 0.02-0.05 m; 2: 0.05-0.10 m; and 3: >0.10 m to create the surface index. A lower index indicated less damage due to trampling. Visual evaluation was also performed using photos. The documentation was performed once before the grazing season started, three to four times during grazing season (depending on rotation intensity), and once after the grazing season ended. The number of cow passes through the entrances was documented continuously. Temperature and precipitation were documented continuously at the field experiment using a local weather station. Statistical analysis was then performed, where the surface-index was response variable (Y), and treatment and documentation occasion were explanatory variables (X) (SAS, 2008).

Results and discussion

The first grazing season was dryer than normal for the site. The second grazing season had normal precipitation (except for a dryer July), resulting in different conditions between years concerning risk of trampling damage. None of the years or parts of the two seasons could be considered as having wet conditions.

The number of cow passes differed between years and between the three blocks due to location and paddock differences in biomass production, which influenced the rotation intensity (Table 1). In the field trial we focused on achieving similar number of passes within each block. There was no significant correlation between the number of passes and surface index (P<0.05) for either of the two years.

Both years the bark chip bed had more trampling damage and a significantly higher surface index than the armoured mats and the control (Figure 1). In the first year one plot with bark chips lost its bearing capacity (collapsed) due to the highest number of cow passes, i.e. 7,283 passes. During the second year, with normal precipitation, all three plots with bark chips collapsed. The visible evaluation showed that the cows often avoided walking over the bark chip beds and instead chose to walk on a thin passage beside the beds. As this comparison has been performed on clay soil it is, however, possible that bark chips perform better on other soils. The control area where no action was taken had some trampling damage and a low surface index (Figure 1). In this rotational grazing system where biomass had the chance to re-establish between grazing activities in the control area, and in combination with good growing conditions, the measure of taking no action was sufficient to avoid trampling damages under the prevailing dry to normal weather conditions during the two years. In the second year the control area had a similar surface index to the armoured mats (Figure 1).The armoured mats had no visible trampling damages and a low surface index for both experimental years. So far the armoured mats have been able to withstand two grazing periods and is a promising stabilizing material. Investment costs for 100 m^2 armoured mat was 21,900 SEK and for 100 m^2 bark chip bed was 6,500 SEK (Nilsson, 2013). Including costs for maintenance and calculating with a life span of ten years the armoured mats need to withstand seven years to reach the same cost as the bark chip beds.

The surface index method was not sufficient when comparing degree of trampling damage between the two stabilizing materials (bark chips and armoured mats). The bark chip material is characterized by having an uneven surface, which resulted in a more than five times higher surface index than the armoured mats and control at the first documentation occasion before entrance of the cows. Complementing with

Table 1. Average number of cow passes documented during the grazing season

Year	Bark chips	Armoured mat	Control
1	4,810	5,070	2,500
2	3,780	4,090	3,850

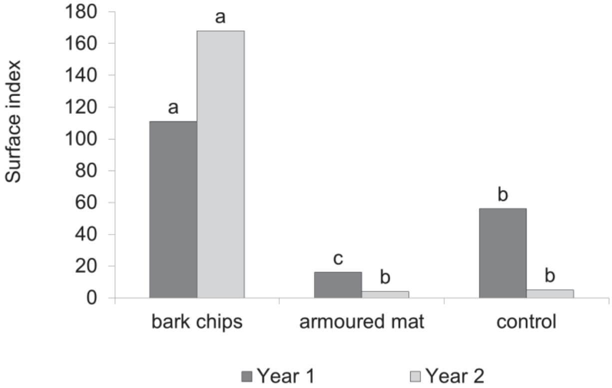

Figure 1. Average surface index with different letters within each year are significant different (P<0.001).

visual evaluation showed clearly the moment when bark chip beds reached a stage with permanently deep holes and when cows avoided to passes over them.

Conclusions

The armoured mats showed the greatest resistance to trampling damages and were hardly affected during two grazing seasons and a winter in between. Calculations showed that the costs of armoured mats can be as low as bark chip beds if they can be used for seven years. The bark chip beds did not withstand trampling under the conditions of this experiment. Despite this, bark chip beds can be a cheap stabilizing material if the farmer has access to bedding material and can replace the bedding surface during the grazing period (Lindgren and Benfalk, 2004). The measure of taking no action on trampled sub areas was sufficient for current dryer weather conditions than normal in this experiment with a rotational grazing system.

Acknowledgements

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Protein productivity and extractability of legume and grass species during spring growth

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Abstract

Legume and grass crops are potential alternative protein sources to substitute the high levels of importation of soybean meal for animal production. The quantities of crude protein fractions were estimated in legumes, namely white clover (*Trifolium repens* L.), red clover (*Trifolium pratense* L.) and lucerne (*Medicago sativa* L.) and grass species, namely perennial ryegrass (*Lolium perenne* L.) and tall fescue (*Festuca arundinacea* L.) across six harvest dates during spring growth, using the Cornell net carbohydrate and protein system. The production of crude protein fractions was strongly affected by species. Red clover showed the highest B_3 fraction productivity and true protein extractability across the harvest dates compared with other species. White clover and red clover demonstrated higher production of B_2 fraction in early growth compared with other species.

Keywords: CNCPS, protein fractions, spring growth, morphological development, legume, grass

Introduction

European countries are importing large quantities of protein mainly in the form of soybean meal for animal production. The extent of feed proteins imported into the EU has raised various concerns such as susceptibility of animal production to price volatility, deforestation, biodiversity loss, and soil and water pollution (Van Krimpen *et al.*, 2013) which indicates the importance of identifying alternative protein sources within the EU to substitute for the importation of soybean. The quantity, quality and price of the alternative protein should be comparable to that of soybean meal if substitution is to be successful. Legumes and grasses are important sources of protein due to the high quantity of up to 250 g kg^{-1} dry matter (DM) (Lyttelton, 1973). Moreover, the nutritional quality of leaf protein was shown to be comparable to that of soy protein (Pirie, 1987). If leaf protein production can be integrated with a biorefinery which produces other products, the overall economics could be considerably improved (Dale *et al.*, 2009). Therefore, biorefinery of grasses and legume crops for production of protein concentrates for animals is an interesting option. In this study, protein fractions were estimated according to the Cornell Net Carbohydrate and Protein System (CNCPS) which fractionates the crude protein into five different fractions. It was assumed that the protein fractions estimated by CNCPS correlate with the extractability of protein fractions in a biorefinery. The objective of this study was to estimate the protein fractions of legume and grass species during the spring growth.

Materials and methods

The legume (white clover, red clover, lucerne) and grass (perennial ryegrass and tall fescue) species were harvested during the spring growth on 6 different dates: 11 May, 19 May, 26 May, 1 June, 8 June and 16 June. Plots were established in 2014 and harvested in 2015. Grasses were fertilized in spring 2015 with 140 kg N ha^{-1} and legumes were unfertilized. Field plots were arranged according to a split plot design with four replications. Samples were analyzed for crude protein fractions according to CNCPS into five different fractions using the methodology of Licitra *et al.* (1998). Fraction A was estimated using trichloroacetic acid (TCA) as protein precipitant agent and determination of total Kjeldahl nitrogen in the precipitate. Crude protein fraction B_1 was determined as borate-phosphate buffer insoluble protein fraction subtracted from TCA precipitated protein. Fraction B_3 was assessed as acid detergent insoluble protein (ADIP) deducted from neutral detergent insoluble protein (NDIP). Fraction C was estimated as ADIP, and finally fraction B_2 was estimated by deducting NDIP from the buffer insoluble protein fraction. Effect of species and morphological development on the quantity of protein fractions were evaluated according to the mixed model using R statistical software. Plant species and morphological development were fixed variables while field replication was random variable. Pairwise comparisons were performed using the Tukey HSD. A probability of $P<0.05$ was declared significant.

Results and discussion

The amount of each protein fraction of legume and grass species across harvest dates is presented in Figure 1. Plant species and morphological development influenced the productivity of crude protein fractions. Assuming fraction C is not extractable and fraction B is extractable, the highest extractable true protein ($B_1 + B_2 + B_3$) was determined for red clover at most of the harvest dates. This could be attributed to the significantly higher *($P<0.05$)* amount of B_3 fraction compared with other species. The highest extractable true protein for red clover was achieved at the 4th harvest date with a value of 726 kg ha^{-1} after which there was a decline in the amount of extractable true protein. At the second harvest date, white clover demonstrated extractable true protein content (404 kg ha^{-1}) which was significantly *($P<0.05$)* higher than that of all other species except red clover. The highest extractable true protein content for white clover was achieved at the 5th harvest date with the value of 535 kg ha^{-1}. Grass species contained the lowest extractable true protein at the first harvest date and the value increased *($P<0.05$)* from first to the last harvest date. Extractable true protein increased from 221 to 478 kg ha^{-1} for perennial ryegrass and from 202 to 449 kg ha^{-1} for tall fescue from first to the last harvest date.

The extractability of true protein, meaning $B_1 + B_2 + B_3$ was based on the assumption that B_3 fraction which is bound to the cell wall might become extractable using enzymes. However, if B_3 is not extractable, true protein can be defined as $B_1 + B_2$ fractions which was shown to be significantly higher *($P<0.05$)*

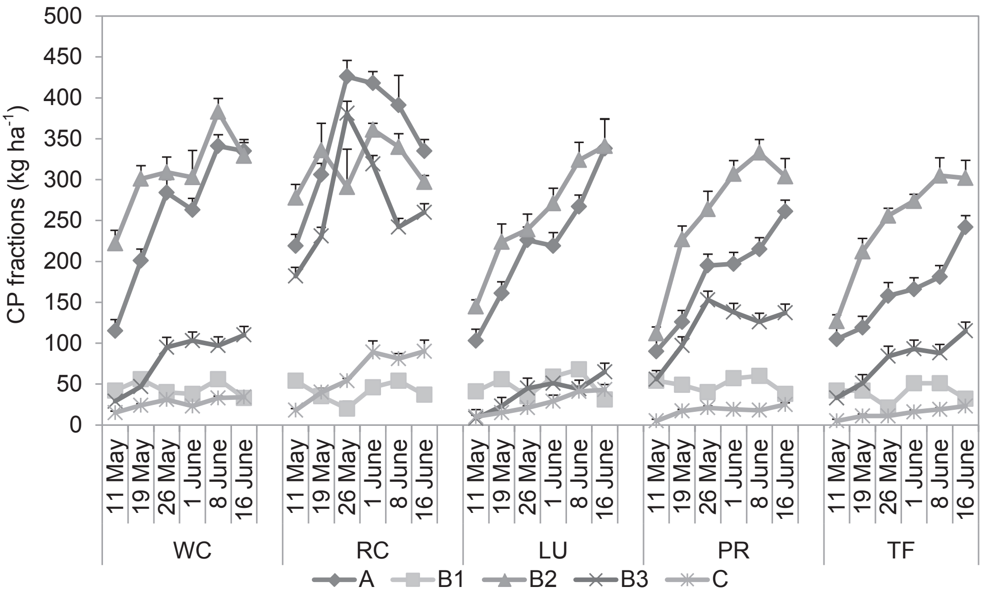

Figure 1. Amount of individual CP fractions (kg ha^{-1}) for legume (WC= white clover, RC= red clover, LU= lucerne) and grass (PR= perennial ryegrass, and TF= tall fescue) species across the harvest dates during the spring growth. Bars indicate the standard error of the mean. Error bars that are smaller than the width of the symbol are not visible.

for white clover (265 kg ha^{-1}) and red clover (332 kg ha^{-1}) only at the first harvest date (early growth) compared with other species. McKenzie (1977) reported protein yields of 75 and 77 kg ha^{-1} on May 2 and June 6 respectively for white clover which was achieved using a laboratory scale pulper and press. Irrespective of the fact that our results only represent an estimation of protein extractability in a biorefinery, the discrepancy in the quantity of extractable protein (kg ha^{-}1) can be attributed to differences in the DM yield and protein extraction condition in general; as alkaline pH, warm aqueous extraction solutions and pretreatment (disruption of cell walls) benefit the protein extraction (Dale *et al.*, 2009) which were not incorporated in McKenzie's (1977) study.

Generally, the B_2 fraction exhibited the highest and fraction C the lowest amount in all species. There was no significant difference in the quantity of fraction B_2 between grass species at any harvest date. Moreover, grasses and legumes did not differ significantly in B_2 fraction content at the 3rd and last harvest dates. However, the amount of B_2 fraction was higher ($P<0.05$) for white clover and red clover at the first two harvest dates compared with other species. The amount of fraction B_1 did not differ among the species at any harvest date.

Conclusions

The results showed that the amount of crude protein fractions can be manipulated by selection of both morphological development stage and plant species. We anticipate that the estimation of crude protein fractions can provide knowledge of the suitability of the crops for protein production in an integrated biorefinery system. However, it is not yet determined how well the actual extractability of different crude protein fractions correlates with CNCPS crude protein estimation. This will require further investigation of parallel physical extraction and CNCPS estimation.

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Drawing pathways of cattle farms to identify the factors of grassland maintenance in the long term

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Abstract

French agriculture has been facing a renewal of interest for grassland since the nineties due to their multiple functions. Nevertheless, grasslands have diminished in French cattle territories as a result of agricultural intensification and homogenization. However in some local areas a maintenance or increase of grassland has been observed during the last decades. By questioning farmers in one of those areas (Brittany, France), we aimed to identify and analyze the changes of farming systems leading to grassland maintenance in the long term. The qualitative surveys were led on the whole farmer career to collect data enabling the analysis of: (1) the farm strategy pathway and (2) the changes in grassland use to reach the strategy. The results show 6 different strategy pathways related to two axes: (1) specialization vs diversification and (2) intensification vs agro-ecological management. The focus on the grassland use showed 4 kinds of grassland pathways related to 6 aggregated indicators of grassland roles: the quantitative place of grassland in the farming system, its zootechnical role, the flexibility and security it brings, the agronomic and environmental roles. Our work shows that grassland maintenance is mainly supported by the feeding roles of grassland and occurred in various strategy pathways and partially determined by the strategy pathway. Our work is important to imagine new ways to help the grassland maintenance on farm according to the farmer's strategy and the role grassland can play to reach it.

Keywords: grassland, farming system, pathways

Introduction

In France, grassland areas have decreased since 1960 due to specialization and intensification of cattle production. However, research programs have shown the benefits of grasslands use (natural/permanent/temporary) such as: (1) better incomes at farm scale or territory scale; (2) environmental; (3) and social benefits (Huyghe *et al.*, 2014). In spite of public policies and technical tools promoting grassland use, grassland areas are still decreasing, mainly in plain regions where cattle production is dominant. Nevertheless a recent study led on the two last French agricultural census datasets (2000 and 2010) highlighted that local dynamics of grassland maintenance exist over the period among the general decrease of grassland areas (Couvreur *et al.*, 2016). Knowledge about how grasslands maintained at farm scale in this region could help stakeholders in developing technical tools and advice for grassland development. For this purpose we aimed at identifying and analysing the changes of farming systems that lead to grassland maintenance in the long term. Our hypothesis was that maintenance of grassland is linked to the farm strategy pathway and the roles grasslands play for the forage system and have for agronomical and environmental benefits.

Materials and methods

The study took place in the nearby area of Rennes (Brittany, France). In 2015, we studied 12 dairy and/or beef cattle farms selected to be representative of the diversity of farming systems in the area according to the productions and the farms' size. The farmers had to have been settled before year 2000. We collected the data by one-to-one survey performed twice with the farmers knew the system best and its historical evolution. The data were collected, from the time of settling.

The first interview dealt with the structural evolution of the farming system (utilized agricultural area (UAA), work force, production facilities) and dimensions of the different crop and animal productions (production volume, number of animals, crops rotations). Singular events like cessation, creation or expansion of a production workshop, major changes in the crop and forage systems management, structural evolution made possible the drawing of farm strategy pathways. Each farm is characterized by stability periods and changes. A stability period represents a time period while there is no major change in the production strategy.

The second interview focused on grassland management during each stability period by questioning about the type of grasslands, type of forage, grass forages quality and quantity, grazing management and crops rotations including grassland. Then we characterized the grassland place within each period of the strategy. Grassland place was assessed according to 6 aggregated indicators judging its potential roles: (1) quantitative place of grassland in the system (QPG); (2) zootechnical role; (3) forage system security; (4) forage system flexibility; (5) agronomic services and (6) environmental services. Each indicator corresponds to the sum of weighted criteria. A criterion results from the coding of one variable (e.g.: part of grassland in UAA, percentage of sock based on grassland, number of type of grassland storage, number of different type of grassland…). The coding and the weight for each criterion have been established regarding to the literature (Baumont *et al.*, 2009; Delaby and Peyraud, 2009; Delagarde *et al.*, 2001). Then by analyzing the evolution of those indicators, we split farms into grassland pathway groups. Finally we analysed the relation between farm strategy pathway and grassland pathway to identify the main factors of maintenance of grassland at farm scale.

Results

We highlighted 7 strategy pathways for the 12 dairy farms surveyed according to two dimensions: (1) specialization vs diversification and (2) intensification vs agro ecological management. The pathways are: (1) organic specialization (n=1); (2) sustainable specialization (n=2); (3) organic diversification (n=1); (4) intensive specialization in dairy or beef cattle production (n=3); (5) diversification with a monogastric workshop (n=2); (6) diversification with a ruminant workshop (n=2); (7) diversification with cheese making (n=1).

We highlighted 4 types of grassland pathways regarding to the 6 indicators (Figure 1). For 3 farms, all the indicators decrease and constitute G1 group. QPG and security increased in 9 farms (G2 and G3). G2 group (n=1) differed from G3 group (n=8) in higher agronomical and environmental roles and lower flexibility and zootechnical roles. G3 group can be divided in 2 sub-groups corresponding to the behavior of the pathways: regular pathways maintaining the same direction (G3_1, n=6) and broken pathways indicating a brutal change in the grassland use (G3_2, n=2).

When comparing grassland and strategy pathways, we observed that grassland roles were not explained by the farmer's strategy. For example G1 group gathers 3 different types of strategy pathways and G3 group gathers 7 different types. Nevertheless, when grassland roles increase (G3 group), the way they increase (efficiency, substitution, reconception) seemed related to the farm strategy pathway. Thus the sustainable specialization pathways are characterized by two separate steps: strong increase of QPG and security followed by an increase of flexibility and zootechnical roles. The two farms with a strategy pathway going to organic farming also had similar grassland pathway with linear increase of QPG, security, flexibility and zootechnical roles of grassland. Finally, diversification with a monogastric workshop strategy pathway is mainly characterized by a steady increase of the flexibility and zootechnical roles.

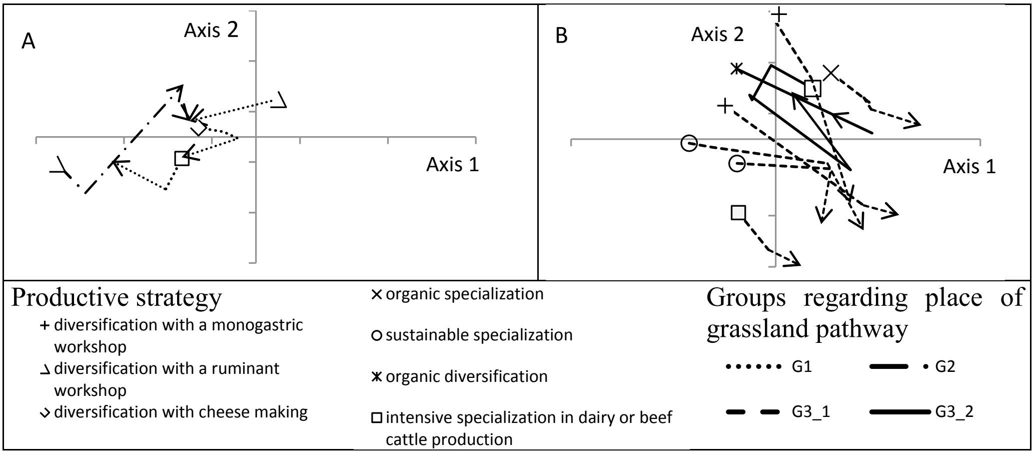

Figure 1. Place of grassland in the various productive strategy pathway of the farming system projected on two axes (A) G1 and G2 groups; (B) G3_1 and G3_2 groups. G1: decreasing place of grasslands pathway for all indicators; G2: increasing quantitative place of grassland in the system (QPG), security, agronomical and environmental roles of grasslands; G3_1: increasing QPG, security, flexibility and zootechnical roles of grasslands; G3_2: increasing QPG, security, flexibility and zootechnical roles of grasslands with brutal changes in grassland use. Axis 1: mainly positively related to QPG and security role and secondary to flexibility of the forage system, zootechnical, environmental and agronomical roles. Axis 2: opposes at the upper part agronomical and environmental roles and on the opposite flexibility and zootechnical roles.

Discussion

Our work shows that, in the studied population, room for manoeuvre was mobilized to increase grassland use at the farm scale independently from farm strategies. As they were mainly focused on the zootechnical and forage system flexibility, the grassland roles for feeding management seem to be the main factor of maintenance. This is certainly linked to the fact that ruminant production was the major farm workshop. Whatever the starting point, grassland pathways are mainly evolving toward an increase in the place of grassland in the farming systems, partially linked to the farm strategies. From that statement, we can make the hypothesis that there are other factors than farm strategy that affect the design of the forage system and contribute to the maintenance of grassland at the farm scale. One of these factors can be the socio-economic context (stakeholders' territory, agro-food production chain) which influences farmers' points of view about grassland use (Geels and Schot, 2007; Gibon, 2005). An issue for the future is to characterize the impact of the socio-economic context on grassland use, in order to enhance its use at the farm scale. For that purpose a second part of the study will focus on the influence of networks (professional or not) on farmer practices.

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Local spatio-temporal dynamics of grassland maintenance between 2000 and 2010 in French cattle areas

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Abstract

The area and proportion of grasslands in the French agricultural area are decreasing, mainly in regions where cattle farming is dominant. Our aim was to study if local dynamics of grassland maintenance exist in those regions. Data taken from the two last agricultural censuses 2000 and 2010 at the smallest French administrative scale ('canton') were used. 977 cantons were selected to represent those intensive cattle farming regions. A spatio-temporal statistical analysis allowed us to identify 4 profiles of grassland maintenance dynamics: (1) an increase in area and proportion of grassland in a cluster composed by at least 5 cantons; (2) an increase or maintenance of area and proportion of grassland in one canton while a decrease in its neighborhood; (3) a decrease of grassland area but an increase of its proportion in the agricultural area in one canton, and a decrease in its neighbourhood; (4) a decrease in area and proportion of grassland in one canton but lower than its neighbourhood. Depending on location, the local dynamics of grassland identified were linked to complex combinations of factors (pedoclimatic conditions, stakeholders, urban areas…) where public policy appeared as a common explanatory factor for grassland maintenance in numerous areas.

Keywords: grassland maintenance, spatio-temporal dynamics, cattle farming

Introduction

Despite a decrease in French grassland areas between 1960 and 2005, research programs have shown that grasslands (natural/permanent/temporary), associated or not to other forage crops, can offer multifunctional services improving (1) economic (Garambois and Devienne, 2012), (2) environmental (Hopkins and Holdz, 2006), and social farming system sustainability (Huyghe *et al.*, 2014). Based on these results, public policies as well as technical tools and advices have been developed since the end of the 90's to promote the development/ maintenance of grasslands in cattle farming systems. Nevertheless, grassland areas still decreased during the last 10 years (-2%), mainly in the western regions where cattle farming is intensive and dominant. This decrease is described in the literature at a large scale (regional). As there is a high diversity of crop systems at a local scale, our hypothesis was that despite a general decrease in cattle areas, local spatio-temporal dynamics of increase/maintenance of grasslands exists. The objective of our study was to analyse the local spatio-temporal dynamics of grassland areas, between 2000 and 2010, in regions where cattle production is dominant and grassland areas is low in comparison to the French average.

Materials and methods

We used the two last French agricultural censuses dataset (2000 and 2010) at the 'canton' scale which is a local administrative unit (which represent 3,664 cantons in the metropolitan France). We considered that a 'canton' was a part of a cattle area if: (1) the proportion of the utilised agricultural area (UAA) dedicated to cattle was higher than the national average of 42% or (2) the share of cattle farms was higher than the national average of 34% or (3) the number of cattle was higher than the national average of 5,231 heads. Then, we calculated the productive grassland (PG) area which is the sum of the temporary grassland area and the productive permanent grassland. In the selected 'cattle cantons', we considered a 'canton' with little grassland areas if it had a proportion of PG in the UAA lower than the average of 50.56%. This made it possible to eliminate mountain regions where permanent grasslands predominate and cattle farming system is mainly extensive. We selected 977 'cantons'. Several cantons were isolated and others constitute continued areas in the North-West (NW1 and NW2), North (N) and East parts of France.

We focused our work in the NW1, NW2 and N continued areas to detect the spatial structure and dynamics of grassland location. Proportion of PG and rates of evolution between 2000 and 2010 of PG area and proportion of PG were used as input data. In order to gain insight into how grassland proportion changes across region, the local spatial autocorrelation using local indicators of spatial association (LISA indicator, the local Moran's statistics) was calculated (Anselin 1995). Those statistics provide a measure, for each spatial unit (canton) in the region, of the unit's tendency to have a value (of grassland area or proportion) that is correlated with values in nearby areas (similar or dissimilar values) (Anselin 1995). The objective was to determine some cantons where grassland is maintained or developed between 2000 and 2010 and where dynamics are either similar between neighbours (clusters) or different from the neighbourhood (outliers). The high-high (HH) and low-low (LL) locations were referred to as spatial clusters, while the high-low (HL) and low-high (LH) locations were termed spatial outliers.

Finally, to understand the global dynamic of a geographical area, we surveyed stakeholders (advisors, experts, facilitators) and farmers in 7 clusters characterized by a positive PG evolution. Interviews were conducted according to 5 factors supposed to influence PG evolution: pedo-climatical conditions, socio-professional context, environmental issues, local policies, and economical context. We aimed to classify the factors for each cluster to identify the one appearing the most important in spatio-temporal PG positive evolution.

Results and discussion

We observed HH, LL, LH and HL dynamics in the studied regions. LL clusters and LH outliers were more important and extended (Figure 1). This result showed that a lot of 'cantons' lost grassland area, confirming an erosion of grassland areas at a regional scale between 2000 and 2010. This decrease is related to specialization and intensification of milk production, due to increases in maize crop areas at the expense of PG (Peyraud *et al.*, 2009). In England, grasslands has increased in cattle areas in relation to the development of beef cattle farming and/or optimal pedo-climatical conditions for grassland production (Fowell, 2010).

Local dynamics of grassland maintenance exist. Continuous HH clusters were found in the three studied regions. The HL outliers were mainly located in the NW1 and NW2 regions. Even if the method used does not give explaining factors for grassland evolution, it gives original results that show how it could be interesting to associate spatio-temporal statistical analysis at lower scales to go beyond results and interpretations from larger scales. We observed 4 profiles of grassland spatio-temporal dynamics (HH clusters and HL outliers): (1) an increase in PG area and proportion of PG in the 'canton' and its neighbourhood; (2) an increase or maintenance in PG area and proportion of PG in the 'canton' while a decrease for these two variables in its neighbourhood; (3) a decrease in the PG area but an increase in the proportion of PG in the 'canton', and a decrease for these two variables in its neighbourhood; (4) a decrease in PG and proportion of PG in the 'canton' but lower than its neighbourhood.

PG could be locally maintained in cattle breeding areas despite a general trend of specialization, intensification, and herd enlargement which mainly lead to an increase of maize as principal forage for cattle diet. Our surveys showed that grasslands are conserved due to a complex combination of factors (pedo-climatic conditions, stakeholders, urban areas…) even if the public policy appeared as a common reason for grassland maintenance in numerous areas. In wetlands, environmental policy tools take part in grassland maintenance by supporting land use and landscape preservation. Those measures illustrate

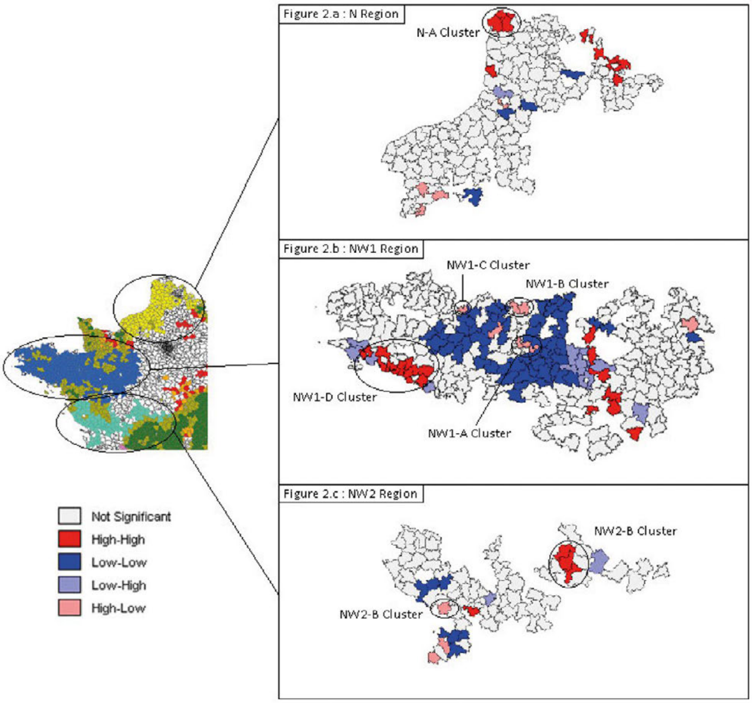

Figure 1. Spatio-temporal dynamics of the proportion of productive grassland areas, between 2000 and 2010, in regions where cattle production is dominant and grassland areas are low in comparison to the French average. (N: North region; NW1: NorthWest region in Brittany and north part of Pays de Loire Regions; NW2: South part of Pays de Loire, North of Poitou and West of Centre Regions; A, B, C, D ... Examples of cantons within a region with a positive spatial grassland dynamic).

that society recognizes the role of wet grasslands in the preservation of biodiversity and landscapes, and therefore the multi-functionality of agriculture. In this case, public policies are the pillar of grassland maintenance which constitute a worrying situation in case of its suppression. In other areas, maintenance of grassland results from the complex combination of several dynamics (farmers looking for ways to reduce feeding costs, innovative management of grassland, alternative forms of advice, increasing interaction between farms and cities leading to new ways of production,...). The hypothesis is that grassland crystallizes several issues and stakeholder's strategies for land use which finally contribute to its maintenance. Grassland maintenance results from the balance between actions of the different stakeholders of territories: farmers, policy makers, civil society (Gibon, 2005).

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Grazing practices, perception and expectations of Walloon dairy farmers

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Abstract

The role of grasslands as a C sink is generally accepted. It is considered that permanent grasslands allow annual C storage rates between 22 and 44 g C m^{-2} y^{-1} (Soussana *et al.*, 2010) thereby contributing to the mitigation of greenhouse gas emissions. Grassland preservation has several other advantages including a decrease in feeding costs (Dillon *et al.*, 2005), a positive effect on cows' health (e.g. a decrease in lameness) (Burow *et al.*, 2011) and the provision of a positive image to consumers. Despite these arguments, grazing is decreasing in Europe and grasslands are disappearing. A better understanding of grazing practices and of farmers' expectations could suggest ways of improving these practices and limiting grassland disappearance. For this reason, a survey was conducted in December 2015 amongst Walloon dairy farmers and the preliminary results are presented below.

Keywords: permanent grasslands, grazing, dairy industry, dairy cattle

Introduction

Permanent grassland is considered to play a role in mitigating greenhouse gas emissions by acting as a sink for carbon. Studies have estimated that the annual C storage rates are between 22 and 44 g C m^{-2} y^{-1} (Soussana *et al.*, 2010). Moreover, the use of grassland for grazing seems profitable because it brings about a decrease in feeding costs (Dillon *et al.*, 2005). It also has a beneficial effect on cows' health (Burow *et al.*, 2011) and is viewed positively by consumers.

However, an increase in herd size and the automation of herd management have brought about a decrease in the extent of grazing and grassland area. The purpose of the Life-Dairyclim project funded by the European Commission is to describe grazing practices in the three partner countries and to monitor changes in land use. This paper presents the results from Wallonia.

Materials and methods

Eighteen questions about grazing were formulated, focusing on the description of the farms, on grazing practices and on farmer perceptions and expectations. In Belgium, hard copies of questionnaires were sent to 3,152 Walloon dairy producers by the Comité du Lait on 10 December 2015. Questionnaires were also available on the internet.

Results and discussion

A total of 965 completed questionnaire forms were returned, representing a 31% response. Of these, 90.5% came from conventional farms and 9.5% from organic farms. These proportions are similar to those of the two systems in Belgium (Anonymous$_A$, 2015). A little over a third (36.4%) of the farms specialised in dairy production, 22% produced meat and milk, 13.5% produced milk and cereals and 27.9% produced meat, milk and crops. 60% of the farmers were more than 50 years old and 78% of the farms were owned by one person.

The agricultural area on 76% of the farms was less than 100 ha and 87% of the farms had less than 100 dairy cows. Milk production was below 8,000 litres per cow annually in 68% of the conventional farms and 99% of the organic farms. The producers with more than 12,000 litres represented around 1% of the farms. Holstein cows were the most common breed with an average of 78% (81% in conventional and 57% in organic farms). These figures correspond to the average farm structure in Belgium (Anonymous_B, 2015).

Grazing practice is common in Belgium (96%). In the conventional system, 79% of young animals, 98% of heifers, 96% of dairy cows and 90% of dry cows were reported as grazing. In organic farms, the proportions were as follows: young animals: 92%, heifers: 99%, dairy cows: 100% and dry cows: 97%. Compared with zero-grazing farms, grazing ones were smaller in herd size (Figure 1) and surface: only 2% of them had more than 150 cows, vs 32% (10/32) for zero-grazing herds and only 10% of grazing farms use more than 125 ha vs 25% in zero-grazing ones. Large herd size thus seems to hinder grazing.

The length of the grazing season was usually 4 months or more (96%). Most farms grazed the lactating cows both day and night (74%). Supplement feed was given to the lactating cows in 99% of the farms during the grazing period. Moreover, 74.5% of the farms used supplement all the time on conventional farms, vs 37% on organic farms. Maize silage and concentrate mix were the most frequently used supplements during the grazing season (Figure 2). Due to this high level of supplementation, the intake of grass in the summer period was evaluated at less than 50% of the total dry matter intake (TDMI) in 43% of conventional herds. Only 3.5% of farmers estimated an intake of grass of more than 75% of TDMI. On 9% of the organic farms, cows did not receive any supplement at all during the summer and 88% of the farms estimated an intake of grass of more than 50% of TDMI.

The opinions about grazing depended on the type of farm. Conventional farmers considered that grazing decreased production costs (76.8% of the farmers), was beneficial for landscape (81.8%), increased animal welfare (94%) and was positive for the environment (73.7%). Their expectations were to increase grazing at 42.2% of the farms and to keep it constant (39.9%), whilst 4.5% wanted to stop grazing and 13.4% to decrease this practice. There was greater consensus among the organic farmers: 92.1% considered that grazing decreased production costs, 99% that it was beneficial for animal welfare, 91.9% that it had a

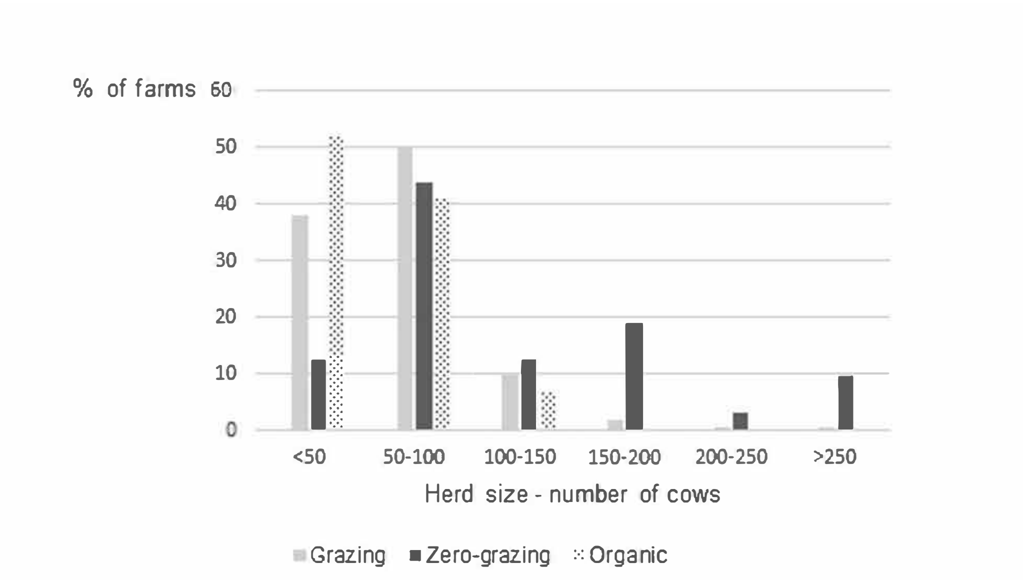

Figure 1. Number of cows per farm on different types of farm.

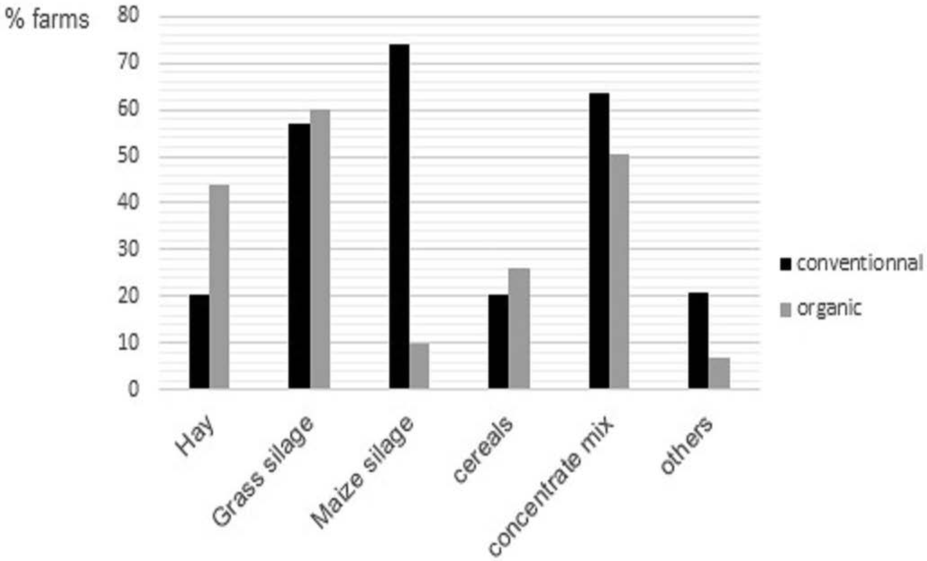

Figure 2. Type of supplementation allocated to the cows during the grazing season.

beneficial impact on both landscape and environment (88.3%). For organic dairy farmers, grazing was expected to be increased (56.3%) or kept constant (38.7%). Only 3.8% of the organic farmers considered stopping grazing.

Conclusions

More than 30% of the dairy farmers answered the questionnaire. Although farmers were very positive about grazing and expected to continue with the practice, the proportion of grass in cows' feed was moderate, even during the grazing period, and a high level of supplementation of grazing cows was reported. This may suggest low confidence in grass quality and quantity. Despite a limited economic benefit due to the high supplementation level, the reasons given for retaining grazing were the reduced feeding costs and improved cow welfare.

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Effects of grassland and grazing management on fatty acid intake and milk – a review

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Abstract

Grazing is important for fresh forage intake, animal health and natural behaviour, landscape values and grassland biodiversity. Grazing is a political, societal and marketing issue in the Netherlands where retailers sell dairy products labelled as 'pasture-milk' and dairies pay a premium to those farmers who let their cows have access to pasture. Grazing affects green leaf intake because animals can select. Grazing animals walk outdoors and harvest fresh herbage from a three-dimensional canopy. Botanical composition and grassland management in various farming systems affect herbage intake and milk fatty acid (FA) composition. Grazing animals encounter a vertical gradient while grazing down a sward. Responses in milk FA composition are discussed in a review paper.

Keywords: analytical constraints, grazing management, herbage intake, milk fatty acid composition

Introduction

Change is needed in the design and management of our food systems in order to enhance natural resource use. The increasing interest of consumers in the environment, animal welfare and the origin and method of food production is reflected in growing preference for food products of pasture-based systems of production. Grazed grass is the most economical means of feeding dairy cows. Milk fatty acid (FA) composition is of great interest as dairy products are a substantial source of FA in the human diet. As the FA profile in dairy product fats is comparable to that of the milk fat from which these products are obtained farm milk composition is important. Fat content and FA composition and concentration in cow milk are mainly dependent on the animals' diet. Nutrition therefore constitutes a natural and economical way for farmers to markedly and rapidly modulate the milk FA composition. The amount and quality of herbage consumed and the concentration and fate of FA in feed, digestive tract and mammary gland determine the FA profile in dairy products. Extensive reviews have been published on FA profiles of milk fat (e.g. Chilliard *et al.*, 2007; Elgersma *et al.*, 2006). It has been often found that milk from grazed ruminants has higher levels of polyunsaturated fatty acids (PUFA) than from ruminants that are kept indoors. Grazing-based production systems, including certified organic systems (Butler *et al.*, 2011; Halfa *et al.*, 2013; Schwendel *et al.*, 2015), however do not have exclusive claims to greater levels of PUFA in milk. It is possible to increase such compounds in confinement feeding through the use of supplemental feeds such as oilseeds or fish or algae sources, although this may have undesirable effects on the unsaturated FA composition (Aldai *et al.*, 2013). This paper outlines a review (Elgersma, 2015) on the effects of grazing on the FA composition of milk.

Methodology

There are methodological constraints in determining FA in fresh herbage in relation to collection and/or handling of plant samples, transport conditions and duration after harvesting, storage conditions and duration in the laboratory, extraction procedures and calculation methods (Elgersma, 2015). A meta-analysis of FA concentrations and proportions of alpha-linolenic acid (ALA), linoleic acid (LA), palmitic acid and 'other' FA showed large differences in concentrations of 'other' FA among experiments. It may be caused by differences in analytical equipment or ways of calculation or data presentation and illustrates methodological constraints when comparing data from various trials and laboratories. If the plant matrix of fresh material affects extraction differently compared with wilted materials, then FA concentrations in fresh herbage samples cannot be directly compared with those in wilted, dried or ensiled herbages; this would pose a methodological constraint when comparing processes during grazing compared with indoor feeding.

Sward factors – botanical composition and selection by grazing animals

Green leaf mass is an important parameter because leaf blades have a higher nutritional quality than stems. Grazing animals can select between species and plant parts. Therefore, their ingested diet is usually of higher quality than that of a random sample of the sward. Farmers can maximize opportunities for diet selection by offering grazing animals a high herbage allowance. There are trade-offs between herbage utilization and herbage quality.

Animal selection is both horizontal (between patches, plants and species) and vertical as they start grazing from the top of the canopy in so-called horizons. The mass of herbage within the grazed horizon has a more profound effect on bite mass than sward surface height *per se* (Orr *et al.*, 2001). It is therefore important to maximize the green leaf mass by providing dense, leafy swards to enable the grazing animal to achieve large bite masses. Appropriate choice of grassland species, cultivars and correct grassland and grazing management can help to achieve this.

Effect of grazing on fatty acid intake

Because grazed grass is often younger than grass cut for stall-feeding and certainly younger than grass cut for silage-making, and because grazing animals prefer leaves over stems, intake of FA and proportional intake of ALA under grazing is higher than in indoor-fed cows.

A fundamental difference between grazing and stall-feeding is that grazing animals eat fresh living herbage while cut forage is either living (green chop, with freshness depending on duration and conditions during the time span from cutting until eating), or dead (hay, silage). Furthermore, selection possibilities, i.e. options to reject plant parts of detached material, are minimal; animals can pick up a detached plant part with their mouth but have limited abilities to select the most palatable portion. In contrast, grazing animals can use their tongue and the force of their head. Apart from the more limited option of biting off material once it has been picked up, detached tillers or chops thereof can either be rejected as a whole or have to be ingested as a whole.

The eating and chewing pattern are different for ruminants that have to bite or tear off forage during grazing compared to animals that eat cut forage. With indoor feeding, detached plant parts are often longer than in grazed bites, as forage is usually cut at a stubble height of 4 or 5 cm. After cutting, wilting begins and plants lose water, crispness and rigour. This physical change can affect the chewing pattern and fragmentation of particles of ingested feed boli that are swallowed. It can affect the rumination process and fermentation rate. Khiaosa-ard *et al.* (2015) recognized that, besides selection, differences in intake (i.e. rumen fill) and rumination patterns (Kaufmann *et al.*, 2011) could play a key role in explaining different results found with high-forage DMI in grazing and indoor forage regimens.

Effects of grazing on milk fatty acid

The n-6:n-3 ratio in cow milk essentially describes the concentrations of LA versus ALA as they represent the most abundant n-6 and n-3 FA. A lower n-6:n-3 ratio is therefore indicative of a forage-based diet.

The composition of conjugated FA in the fat of forage-fed ruminants is different from those animals raised under intensive feeding practices. High intake of PUFA from fresh herbage affects rumen fermentation pattern and gives milk fat with less saturated FA and more vaccenic acid, rumenic acid (*cis*-9, *trans*-11 conjugated linoleic acid (CLA)) and ALA compared with indoor feeding (e.g. Larsen *et al.*, 2010). Milk from botanically diverse pastures such as grazed mountain pastures also often contains more PUFA than milk produced on species-poor pasture (e.g. Noni and Batelli, 2008); secondary plant metabolites in forbs may reduce rumen biohydrogenation. Khiaosa-ard *et al.* (2015) concluded from a meta-analysis that grazing regimens led to the strongest enrichment of rumenic and vaccenic acid in milk fat. Surprisingly they found that, when C18 PUFA intake was very limited, their transfer apparently exceeded 100%, assuming that intakes reported in the studies the authors used for their meta-analysis were correct. It is known that estimations of intake are prone to variation, particularly under grazing (e.g. Smit *et al.*, 2005). Moreover, there could be bias in the calculation method when relating milk composition to intake in grazing studies, because diet effects of day 1 are expressed in milk of day 2. As reviewed by Elgersma (2015), in some studies milk composition has been related to the diet of the same instead of the next day, which led to errors in data interpretation. This methodological pitfall is less disturbing with indoor feeding experiments where a constant diet is fed, where it would in fact only have shortened the assumed adaptation period with one day.

Conclusions

As herbage allowance can be managed easily by farmers, it might represent an interesting and feasible way to obtain higher concentrations of FA in herbage. Also decisions on the timing of cutting or grazing, i.e. the regrowth stage and phenological development at which herbage is harvested or cows are allowed to enter the pasture, can be a practical means of influencing herbage quality, FA intake and milk FA profiles.

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BEHARUM project: use of ICT to monitor feeding behaviour in grazing ruminants

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Abstract

Developments in microelectromechanical system (MEMS) and information and communication technologies provide new opportunities for the automatic monitoring of animal behaviour. The BEHARUM project is aimed to realize an automatic recording device for monitoring the grazing behaviour of ruminants. The device includes a small, low power, complete three-axis accelerometer with signal-conditioned voltage outputs. It can measure the static acceleration of gravity in tilt-sensing applications, as well as dynamic acceleration resulting from motion, shock, or vibration. The sensor inserted in MEMS is attached to the lower jaw and, thanks to its miniaturization, has a minimum impact on the animal. The system is able to continuously sample accelerations and to send three values at 1-s intervals to a nearby computer, by a ZigBee wireless module connected to it, or to a remote computer via Global System for Mobile Communications (GSM). Data acquisition and analysis is realized by a software installed in the receiving computer. Calibration and validation of the system is ongoing, comparing acceleration with video-recording data to automatically classify three activities, eating, ruminating and resting in grazing dairy sheep. A data mining approach followed by multivariate analyses procedures, allows automatic classification of the three activities, saving battery charge to lengthen the recording session. The system is upgradable with other sensors such as the global positioning system and it could become a useful tool to effectively drive the management of pastoral resources.

Keywords: accelerometer, grazing, ruminating, resting, multivariate analysis, dairy sheep

Introduction

Monitoring the behaviour of ruminants in near real-time is important to check the living conditions of animals and manage them more effectively in terms of performance, welfare and environmental impact. The growing access to information and communication technologies (ICT) is boosting precision farming that may allow individual livestock management in order to maximise individual animal potential (Moreau *et al.,* 2009).

The miniaturization of electronic components allows their minimum impact on the animal. Moreover, through the use of wireless modules, is possible to send data upon certain distances opening new prospects for studying animal behaviour where access to visual observation is difficult (Brown *et al.*, 2013). The BEHARUM project has contributed to this research area by developing an accelerometer-based recording device able to classify automatically the behaviour of grazing sheep.

Materials and methods

The project was funded by the regional government of Sardinia to promote research on ICT applied to ruminant management. The involved partners were: the department of agriculture of the University of Sassari, the agricultural research agency of Sardinia (AGRIS Sardegna) and a local enterprise (LE, Electronic System).

The BEHARUM device consists of a halter equipped with a three-axial accelerometer sensor positioned under the lower jaw of the sheep. The animal's movements are detected through accelerations measured in the x (longitudinal), y (horizontal) and z (vertical) axis. The sensor is inserted in a micro-electromechanical compact system (MEMS) with lots of on-board peripherals. The acceleration sensor records both accelerations related to changes in the movements of the sheep (dynamic accelerations) and static accelerations (-9.8 m s^{-2}). The microcontroller samples the row acceleration data at a frequency of 62.5 Hz and encode them, through an analog-to-digital converter with a resolution of 8 bits, into levels ranging from 0 to 255, then selects only three acceleration converted values per second and axis. Acceleration converted data are sent (ZigBee wireless system) to a receiver nearby computer equipped with an antenna or to a remote computer through a local server using the GSM services.

Several calibration trials have been conducted at Bonassai experimental farm of AGRIS, located in the NW of Sardinia (40°N, 32°E, 32 m a.s.l). Three Sarda sheep were equipped with the BEHARUM device and acceleration data were recorded as animals were grazing the following different forages: *Medicago sativa* L., *Cichorium intybus* L., *Hedysarum coronarium* L., *Trifolium alexandrinum* L., and *Lolium multiflorum* Lam.. During each trial sheep behaviour was video-recorded by a fixed camera. Acceleration data were acquired with a software developed by the LE and installed in the receiving computer that stored data in a .csv file.

According to Watanabe *et al.* (2008), from the acceleration data, were calculated the mean, the variance and the inverse coefficient of variation (ICV; mean/standard deviation) per minute for each axis. Moreover, the resultant mean, variance and ICV values of the three axes was also calculated. On the basis of the video recording, the behaviour of the animals was classified, at 1-min intervals, into three activities: grazing, ruminating and resting. Resting activity includes sheep lying down, standing or walking in absence of jaw activity. The dataset obtained combining the three activities with the 12 variables concerning acceleration was analysed by using three multivariate statistical techniques: the stepwise discriminant analysis (SDA), to select the best variable subset, the canonical discriminant analysis (CDA), to test the effective separation among the three behaviours, and, finally, the discriminant analysis (DA), to assign minutes to the correct activity.

Results and discussion

Preliminary calibration results of the BEHARUM device are promising for classifying eating, ruminating and resting activities also in small ruminant as dairy sheep, in agreement with other studies (Moreau *et al.*, 2009; Marais *et al.*, 2014), even when data mining procedures has been applied to the raw acceleration dataset. The SDA selected 7 variables over 12 and the subsequent CDA, developed by using those characters, significantly discriminated the three behaviours (Hotelling's test *P*-value <0.0001). The variation explained by the two extracted canonical functions, CAN1 and CAN2, was 0.68 and 0.32, respectively. CAN1 discriminates between grazing and resting whereas CAN2 differentiates the grazing from the ruminating behaviour (Figure 1). Finally, the DA correctly assigned 92.96% of minutes to behavioural activities. More trials should be run in the near future to complete the calibration process and afterward a validation test may be conducted in different environmental conditions. The system is designed to be upgradable with other sensors such as the global positioning system in order to open its use for spatio-temporal behavioural studies.

Conclusions

The BEHARUM project demonstrates that accelerometers and information and communication technologies are useful tools to discriminate behaviour activities of grazing dairy sheep. Additional types of sensor could be added to this device in order to improve the overall classification accuracy and to effectively drive the management of pastoral resources.

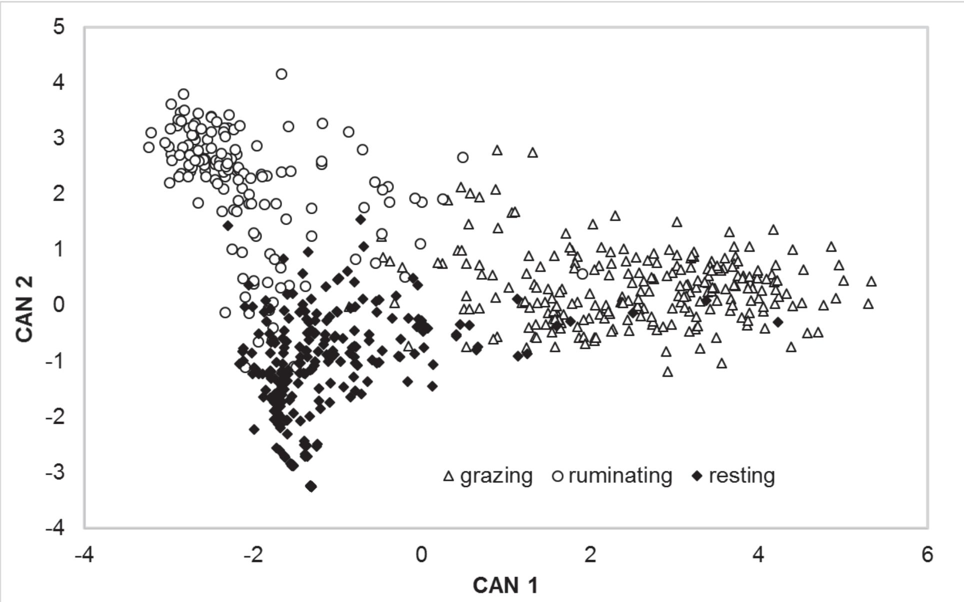

Figure 1. Graph of the two canonical functions (CAN1 and CAN2) obtained in the canonical discriminant analysis applied to acceleration data.

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An analysis of dairy farming and its evolution in Central Switzerland from 2010 to 2014

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Abstract

The autonomy with which Switzerland (CH) pursues its own agricultural policy imposes increased competition. In Central Switzerland (C-CH), 32% of the full-time farms are in the valley region (VR), 20% are in the hill region (HR) and 48% are in the mountain region (MR). In 2014 the average yield of the 3,275 Central Swiss dairy farmers was 7,913 kg of milk (ha and year)$^{-1}$. A full cost analysis carried out from 2010 to 2014 at 421 farms showed that VR farms were more productive than HR and MR farms (75 vs 64 vs 46 kg of milk h^{-1} (working hour) respectively; $P<0.01$). The production (i.e. full) cost was lower in the VR and in the HR than in the MR ($P<0.01$). Labour income was higher in the VR and HR as compared to the MR (15 vs 15 vs 11 € h^{-1}; $P<0.01$). In C-CH, creameries and small holder cheese factories play an important role in creating added value. A promising avenue of adaptation to today's challenging conditions is the prospect of improving the efficiency of the value chain, i.e. by reducing production costs and producing niche products, including specialities (e.g. organics products) and brand merchandising.

Keywords: dairy industry, dairy farming, full costs analyses, productivity, value added chain, sustainability

Introduction

Central-Switzerland (C-CH) consists of six cantons. Forests cover 29.6% of the total C-CH area of 4,483 km^2. Another 40.5% of it is covered by agricultural land, 6.4% is occupied by settlements and 23.6% is categorized as 'unproductive land' (Swiss Federal Statistical Office SFSO, 2015). In 2011, 4.4% of the total working population (or 471,763 individuals) in C-CH was engaged in the primary sector (agriculture, forestry and fishery), 29.1% in the secondary sector (industry and handicrafts) and 66.4% in the tertiary sector (LUSTAT, 2015). In Switzerland, especially in C-CH, the dairy industry plays an important role. The aim of this study was to investigate the evolution of the dairy industry from 2010 to 2014 and to evaluate, in particular, the production costs of dairy farms in C-CH under different geo-economic conditions.

Materials and methods

Data on agriculture, dairy farms and cheese factories were gathered from Milchstatistik (2014, 2010), TSM (2015) and the Central Switzerland Dairy Farmers' Association (ZMP, 2015). Full cost accounting data was processed in accordance with VOKO-Milch + Schweine by AGRIDEA (2014). Statistical analysis was conducted in accordance with the R Core Team (2013). In order to make comparisons, the groups were reported by Bonferroni-Holm adjusted P-values.

Results and discussion

Analysis of farm size and structure: In 2014, 72% of all farms in C-CH were full-time farms. The average agricultural area of these farms (16.7 ha) was lower than the national average (21.2 ha), similar to Austrian and Norwegian farms and lower than the average European holding (Eurostat, 2015). These farms consisted of 86.4% of forage area, mainly grassland, and 11.8% of open arable land. In C-CH, the share of full-time farms in the valley region (VR) was 32%, made up of 4.8% organic farms. In the hill region

(HR), there were 20% full-time farms with 8.2% of them being organic and, in the mountain region (MR), there were 48% full-time farms with 12.5% of them being organic.

Dairy farms: From 2010 to 2014, in Switzerland the number of dairy farms decreased by 29% to 22,597, and milk delivery per ha increased by 6% to 6,089 kg ha^{-1}. The average farm area also increased by 8% to 24.8 ha. In the same period, in C-CH, the number of dairy farms decreased by 11% to 4,796. The yield achieved by the 3,275 ZMP milk producers rose 27% to an average of 147,713 kg (farm and year)$^{-1}$ and by 17% to an average of 7,913 kg of milk (ha and year)$^{-1}$. These numbers reflect the high productive intensity of C-CH dairy farms.

Full cost accounting: Milk yields and labour productivity (kg of milk h^{-1}) varied across all regions (Table 1). More direct payments in the HR and the MR could partly compensate for higher full costs, i.e. overheads (machinery, buildings and equipment) and internal overheads (wage entitlement) in these regions (data not shown). Labour income was significantly higher in the VR and the HR than it was in the MR. Agricultural income from dairy farming was significantly lower in the MR when compared to the VR. In the MR, there was a higher return from by-products (calves or culled cows) as compared to the VR. In the VR and the HR, less than 25% of the dairy farmers earned more than 20 euros h^{-1}. In the MR, less than 10% of the farmers earned more than 20 euros h^{-1}. By calculating earnings at EUR 25.4 h^{-1} for the farm managers, the distribution of profits and losses shows that only about 12.5% of the dairy farms in the VR were profitable businesses, with even lower figures in the HR (10%) and in the MR (5%). In the HR,

Table 1. Full cost analysis of dairy farms (mean ± SEM) in the valley region (VR), hill region (HR) and mountain region (MR) of Switzerland (without group farming).

Bookkeeping data from 2010 to 2014	**VR**		**HR**		**MR**		**Significance**[1]		
About half of these farms are in C-CH[2]	198 dairy farms		87 dairy farms		136 dairy farms		VR vs	VR vs	HR vs
Costs and income per kg milk[3]	mean	±SEM	mean	±SEM	mean	±SEM	HR	MR	MR
Farm size (ha)	29	0.9	25	1.3	22	0.9	*	**	
Number of cows (cows dairy farm^{-1})	35	1.1	28	1.2	21	0.9	**	**	**
Main forage area (are LAU^{-1})[4]	54	0.9	62	1.5	84	2.1	**	**	**
Working hours (labour) (dairy) (h y^{-1})[5]	4,031	83.8	3,847	123.8	3,800	86.2			
Milk sold (kg farm^{-1} y^{-1})	249,720	8,656	186,700	9,478	119,645	5,613	**	**	**
Milk yield (cow $^{-1}$ y^{-1}) (kg)	7,518	79.9	7,076	119.5	6,258	89.6	**	**	**
Labour productivity (kg milk h^{-1})	75	1.6	64	2.1	46	1.3	**	**	**
Milk (main forage area)$^{-1}$ (kg ha^{-1})	12,801	265.9	10,056	340.0	6,821	218.7	**	**	**
Milk price (cent kg^{-1})	57.6	0.53	58.2	0.91	58.7	0.75			
Full costs (cent kg^{-1})[6]	89	1.1	100	2.0	140	3.5	**	**	**
Direct payment (cent kg^{-1})	19	0.4	26	0.8	44	1.5	**	**	**
Profit/loss (cent kg^{-1})[7]	-13	1.0	-16	1.6	-37	2.4		**	**
Labour income (€ h^{-1})	15	0.6	15	0.8	11	0.6		**	**
Agricultural income dairy farming (€ farm^{-1} y^{-1})	47,351	2,526	43,692	3,200	37,451	2,657		*	
from animal by-products (€ farm^{-1} y^{-1})[8]	6,094	460	7,689	680	9,381	803		**	
Agricultural income total (€ farm^{-1} y^{-1})	76,868	3,425	76,437	5,186	57,727	3,396		**	**

[1] *P<0.05; ** P<0.01%.

[2] CH-C = Central Switzerland.

[3] Actual currency 1 CHF = EUR 0.908.

[4] LAU = large animal unit.

[5] y = year.

[6] i.e. direct, overhead and internal overhead costs.

[7] By calculating EUR 25.4 per working hour for the farm manager.

[8] Calves and culled cow returns.

VR and MR, the share of total agricultural income acquired through other on-farm productive activities, such as pig or chicken production, solar electricity, forestry or rented flat, was 43, 38 and 35% respectively.

From 2012 to 2014, the labour income (€ h^{-1}) generated by full-time grazing systems in the VR, HR and MR was higher than that of feeding systems with half-day pasture by 53, 56 and 17%, respectively (Haas and Höltschi, 2015; data not shown). In the VR, organic farms generated labour incomes that were 53% higher than on conventional farms (e.g. with an ecological performance certificate). The labour income on farms with hay conservation systems (for cheese factories) was 25% higher than on farms with silage. In the HR, organic farms and farms with hay generated higher labour income by 47% and 29%, respectively. In the MR, the labour income on organic farms was higher by 18%, whilst on farms with hay conservation systems it was higher by 9%.

Milk processing and cheese factories: In 2014, within CH, 85% of the total milk production (4.1 million t) was processed. In C-CH, 545 million kg of milk was processed, mainly by two large creameries, in the form of cheese (54%), milk preserves (33%) and yoghourt and fresh products (7%). The rest was used for other products. However, the number of small cheese factory cooperatives has decreased from 139 in 2000 to 49 in 2014. Most notably, there was a reduction in hard cheese factories. This precipitous decline in hard cheese factories was due to changes in the behaviour of soft and semi-hard cheese consumers, which was in turn stimulated by the exportation of hard Swiss currency abroad and a shift in farm conservation systems from hay to silage, or the abandonment of milk production altogether in favour of beef production.

Limitations: The farms participating in the full cost analysis were not randomly selected. These data were collected from farms whose managers were attending further vocational training in dairy farming. We suspect that these farms may have an above average performance rate.

Conclusions

Small to medium-sized dairy farms have been placed under considerable strain by high production costs. Productivity in all regions must improve in response to rising competition. For example, farms must shift to a pasture-based system, to organic farming, or to more cooperation, particularly with respect to farm mechanisation and the use of buildings. With increasing demand for meat worldwide, animal by-products will play an important role in the coming years. Furthermore, high returns from by-products and high income resulting from other branches of production will mitigate the income risk posed by high milk price volatility.

If cheese factories can create milk-based specialities and improve their merchandising in the worldwide marketplace (e.g. Emmi Kaltbach Switzerland), they could generate an above average milk price. Such a milk price would partially compensate for higher production costs.

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Milk production and cow behaviour in an automatic milking system with morning and evening pasture access

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Abstract

Legislation in Sweden and Norway requires that dairy cattle have outdoor access in summertime. Pasture utilization can be challenging with high yielding cattle and large herd-sizes. Therefore, many farmers choose to offer their cows access to an exercise- and recreation area only, rather than a full production pasture. However, is an exercise paddock as attractive as production pasture for the cow? We compared part-time production and exercise grazing in an automated milking system, with outdoor access in the morning (4.5 h) and the evening (4 h). The production pasture group (P) was offered fresh production pasture daily (≥15 kg dry matter (DM) cow^{-1}) and given a limited silage ration (6 kg DM) night-time. The exercise pasture group (E) was given access to a small exercise paddock (<1 kg DM cow^{-1}) and were fed silage *ad libitum* 24 hours. Milk yield did not differ significantly: 36.1 kg for P and 36.0 kg for E. However, behaviour differed (P<0.0001), with 5.5 (P) and 2.6 h (E) spent outdoors, and 3.7 (P) and 0.6 h (E) grazing time. In conclusion, while milk-yields were similar between the groups, lower amounts of supplementary feed were needed for cows on treatment P, who also spent longer hours outdoors and grazing.

Keywords: dairy cows, automatic milking, grazing, restricted grazing, supplements

Introduction

Farmers in conventional dairy systems need to fetch the cattle twice a day for milking, but a cow in an automated milking (AM) system may visit the milking unit as she likes within the framework of the system settings. While the latter system may function very well during the non-grazing season, it may offer some challenges when the access area is extended to include the walkways and pasture. Longer walking distances have been shown to affect milking frequencies (MF) and milk yield (MY) (Spörndly and Wredle, 2004), and may cause the cows to act less independently, synchronizing their visits to the AM unit. This leads to an uneven robot utilization and long waiting hours before milking (Ketelaar-de Lauwere *et al.*, 1999). Variation in pasture quality and supply can make an adequate feed supplementation difficult.

Following the introduction of pasture legislation in Norway and Sweden, the use of exercise pasture rather than production pasture has become quite common amongst farmers. The argument is often to fulfil the requirement of having cattle on pasture, while not losing production due to sub-optimal feed supply.

Earlier studies (Ketelaar-de Lauwere *et al.*, 1999; Spörndly *et al.*, 2015) have shown promising results when offering the cows restricted access to pasture, indicating that restricted grazing may be a suitable way to combat the above-mentioned challenges in AM systems. A clear pattern of herd movement between pasture and stable was reported from both studies, and in the latter study a preference of staying indoors during the middle of the day, even when no silage was on offer, was found.

The present study compared production pasture with exercise pasture in an AM system with morning and evening grazing. The hypothesis was that a restricted access to production pasture during morning and evening, with restricted silage supplementation at night-time, would give (1) greater utilization of available outdoor time and (2) higher milk yield than restricted access to exercise pasture with 24 h *ad libitum* silage feeding.

Materials and methods

During a 7 week grazing trial, 41 cows – 22 of the Swedish Red Breed (SR) and 19 Swedish Holstein (SH) – were blocked and randomly assigned into two groups, similarly composed with regard to breed, parity, days in milk (DIM), and milk yield (MY). One group was given access to production pasture (P) and one was given access to exercise pasture (E). A group of non-experimental cows assigned to the P treatment were also included in the herd, to better mimic an AM farming situation with regard to cow numbers.

Treatment P: Cows were offered new pasture daily, at an allowance of ≥15 kg dry matter (DM) cow^{-1}. During night-time, these cows received a restricted grass silage ration (6 kg DM day^{-1}).

Treatment E: Cows had access to the same 0.2 ha field throughout the experiment, with a continuous, low sward height, and low daily allowance (<1 kg DM cow^{-1}), and silage was available *ad libitum* throughout the 24 h period.

A selection gate at the stable exit directed cows to either E or P pasture areas. Cows in both groups were allowed to move freely between the indoor and outdoor areas for a period of 4.5 h in the morning (06-10.30 h), and 4 h in the evening (16-20 h). The remaining time, they were restricted to the stables. Drinking water and supplementary feed was available indoors for both groups. Silage and concentrates were fed and registered on an individual basis (Table 1). Concentrates were fed according to milk production before the experimental start, with rations adjusted every fortnight according the standardised weekly drop in MY (-0.125 kg $week^{-1}$ for primiparous, -0.33 kg $week^{-1}$ for multiparous) used in the Scandinavian feed system 'NorFor'. A commercial concentrate was offered to all cows, and an additional protein-rich supplement for high yielders, Solid 620 and Unik 82 (Lantmännen, Sweden), respectively.

MY, MF, and supplementary feed intake indoors were registered automatically. The grazing behaviour of all cows outdoors was observed for a full outdoor-access period six times during the experiment. For each individual, location (walkway or pasture/paddock), position (standing or lying), and activity (grazing or other) were registered every 15 minutes.

P pasture height was measured daily, and used to calculate new pasture allowance. Samples of pasture and supplementary feed were collected daily for analysis to determine their nutrient composition. The E pasture was mowed at the lowest possible height prior to experiment start and on two subsequent occasions, leaving virtually no grass available for the cows.

The data were analysed in a mixed model using the SAS programme (ver. 9.4; SAS Institute Inc.). The model for analysis of the production parameter (MY) contained the variables treatment (P or E) and breed (SR or SH), using MY prior to experimental start as a covariate. Parity and DIM were excluded due to lack of significance. In analysis of MF and behaviour, the same model was used.

Results and discussion

The metabolisable energy (ME), dry matter (DM), crude protein (CP), and neutral detergent fibre (NDF) contents of the different feed sources are presented in Table 1.

There was no significant difference in the MY between the two experimental groups (Table 2). There were, however, differences in MF; the cows in the E-group visited the AM unit more frequently than did the cows in P-group. The cows in P-group, on the other hand, spent significantly more time outdoors than the E-group. They also spent a greater amount of time grazing while outdoors, and had a higher

Table 1. Mean ± standard error composition (metabolisable energy (ME), dry matter (DM), crude protein (CP), neutral detergent fibre (NDF)) of feed and pasture, sward height, and intake of supplementary feeds for cows on production (P) or exercise (E) pasture.

	Roughages		Concentrates	
	Silage[1]	P-pasture[2]	Basal conc.	Protein conc.
ME (MJ kg^{-1} DM)	11.6 (0.05)	10.7 (0.18)	13.2 (0.00)	14.0 (0.00)
DM (g kg^{-1})	321.0 (0.37)		894.8 (0.18)	889.9 (0.18)
CP (g kg^{-1})	156.9 (0.67)	161.5 (6.04)	177.8 (1.17)	295.5 (1.12)
NDF (g kg^{-1})	425 (4.8)	398 (9.2)	282 (4.9)	243 (0.9)
Sward height (cm)		15.8 (0.52)		
Consumption P (kg DM)	5.6 (0.01)		8.7 (0.08)	1.7 (0.01)
Consumption E (kg DM)	14.6 (0.45)		8.3 (0.09)	1.7 (0.01)

[1] NH_4-N, % of total N: 6.4. Acids, g kg^{-1} DM: lactic acid 75.9, acetic acid 11.8, propionic acid 1.2, butyric acid 0.5, formic acid 1.44.
[2] Exercise pasture was not sampled for chemical analysis as there was virtually no grass available to cows on this pasture.

Table 2. Production and behaviour data (mean ± standard error) for cows on production or exercise pasture.

	Production	Exercise	Sign.
Milk Yield[1] (kg)	36.1 (0.60)	36.0 (0.60)	NS
Milking Frequency[1] (n day^{-1})	2.4 (0.05)	2.7 (0.05)	P=0.0004
Outdoor time[2] (h)	5.5 (0.13)	2.6 (0.12)	P<0.0001
Grazing time[2] (h)	3.7 (0.07)	0.6 (0.07)	P<0.0001
Lying outdoors[2] (h)	1.7 (0.09)	1.4 (0.08)	P=0.0079

[1] Based on daily robot-recordings over 49 days.
[2] Based on 6 observation studies.

total outdoor lying time. However, relative to the total outdoor time, the E-cows spent a greater fraction of it lying than the P-group.

In conclusion, the results support the hypothesis that cows on P-pasture spend more time outdoors and graze longer hours than cows on E-pasture. The data do not support the second hypothesis, however the results show that it is possible to maintain the same MY with a smaller supplementary feed allowance and an increased pasture ration.

Acknowledgements

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I know what you fed last summer – using ^{13}C to retrospectively analyse cattle diets

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Abstract

At most cattle farms, the animals are fed a mixture of grass silage, maize and concentrates in their daily diet; the proportions can vary over the year. Sections from cattle hair contain information on the cattle diet from different time periods. We sampled cattle tail switch hair from 23 cattle farms in northern Germany. Farms differed in feeding (maize, grass, concentrates) and in grazing system (year-round, summer grazing, no grazing). We found that ^{13}C-isotopic signature in cattle hair can be used as an indicator for maize content in cattle diet for a range of production systems. ^{13}C in specific sections of hair showed a high sensitivity to diet changes. We were able to confirm differences in feeding intensity between summer and winter periods as well as short-term diet changes. Our results give evidence that one cattle hair can be used to retrospectively analyse diet compositions for a period of at least one year. ^{13}C in cattle tail switch hair serves as an easy to get, representative indicator for the evaluation of the production intensity of cattle systems.

Keywords: ^{13}C-isotopic signature, cattle hair, maize in diet, production intensity, feeding system

Introduction

As the cultivation of maize often leads to a higher potential risk for larger N emissions compared to grassland forage production (Herrmann, 2013) a larger input of maize is related to a more intensive production. At most cattle farms, especially dairy, the animals are fed a mixture of grass silage, maize and concentrates in their daily diet. The percentages of these components in diet are varying over the year. A simple differentiation between pasture-based and maize-based systems is often not sufficient for evaluating the intensity of the production system. A continuous evaluation with simple and robust indicators is needed (Halberg, 1999). It has been confirmed by isotope ratio mass spectrometry (IRMS) that the isotopic signature of cattle diet is transferred to cattle hair, which serves as an isotopic archive recording dietary changes over time (Schwertl *et al.*, 2003; Auerswald *et al.*, 2011). As the carbon isotopic signatures of C_4 plants, such as maize, strongly differ from that of C_3 plants (grasses, legumes, concentrates like soy or grain), the content of maize in the cattle diet is related to the ^{13}C-signature in cattle hair and can be quantified when calibrated with ^{13}C in diets where the components have been determined (Kelly, 2001). In this study, we analysed the ^{13}C-isotopic signature of different hair sections of tail switch hairs of cattle production systems differing in production intensity in northern Germany. We wanted to find out if ^{13}C can be used to retrospectively detect maize in cattle diet at certain times in the past and thus serve to detect the feeding intensity of cattle production systems during those periods.

Materials and methods

We chose 23 cattle farms (suckler and dairy) in Northern Germany differing in production intensity characterized by the annual milk production per cow and the stocking rate. Farmers gave information on diet composition and grazing systems. On the farms, maize is fed as the only C_4 plant in cattle diet. The farms could be assigned to three grazing systems: year-round grazing, summer grazing (May-October), and no grazing. The diet of these production systems differed substantially in content of maize, grass silage and concentrates (Figure 1).

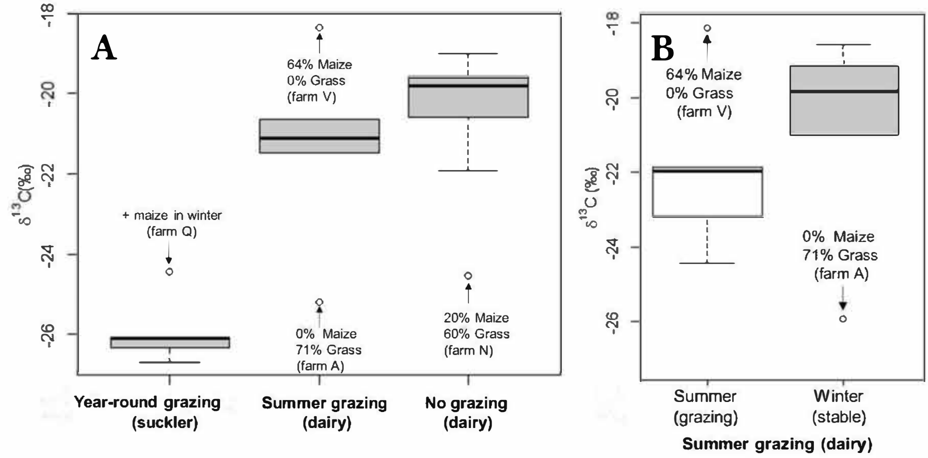

Figure 1 (A). Mean carbon isotope signatures [$\delta^{13}C$ (‰)] for the three production systems and (B) hair sections representing summer and winter period of the summer grazing system of the year 2013.

Sampling of cattle hair was carried out in 2014 and sampled sections contain information on cattle feeding in 2013. We plucked cattle tail switch hairs from two full grown animals of each farm during March 10 and March 20, 2014. All sampled animals had been kept on the farm for more than 2 years before sampling or for their whole life. The hair samples were prepared for stable isotope analysis according to Schwertl *et al.* (2003; 2005). We assumed a medium hair growth rate of 0.8 mm day^{-1} and the isotopic signal of the diet to take 80 days to be clearly detectable in cattle hair (Zazzo *et al.*, 2007; Osorio *et al.*, 2013). Four hair segments of 1 cm each were selected of each hair. For the summer period (August 2013) we chose the section 9-11 cm and for the winter period 2013 (November 2013) the section 3-5 cm. The isotopic analyses was carried out with an isotope ratio mass spectrometer Delta Plus IRMS linked with a Conflo III-Interface (Finnigan MAT, Bremen, Germany) to an elemental analyser NA1110 (Carlo Erba Instruments, Milano, Italy). The isotope data are presented in parts per thousand (‰) as $\delta^{13}C$ (‰), with $\delta^{13}C = [(R_{sample} / R_{standard})-1]\ 10^3$ and R the $^{13}C/^{12}C$ ratio in the sample or standard (V-PDB, 'Vienna Pee Dee Belemnite'). Data were analysed by Kruskal-Wallis rank sum tests and general linear mixed models using the Software R!3.2.2.

Results and discussion

The maize content in diet explained 89% of the variation of $\delta^{13}C$ in cattle hair (P<0.001). A higher content of maize in the diet led to higher (less negative) $\delta^{13}C$. These findings confirm the results of other studies (Schwertl *et al.*, 2003; Auerswald *et al.*, 2011). We found that $\delta^{13}C$ were significantly lower in the year-round grazing system, with no maize in the diet, than in the two other production systems (P<0.01; P<0.001) with 37% (summer grazing) and 41% (no grazing) maize in diet (Figure 1A and Table 1). The $\delta^{13}C$ of the summer grazing system differed between samples from summer and winter (Figure 1B). In this grazing system the content of maize in the diet is noticeably decreased because of a higher intake of fresh substrate from pasture during summer. However, there were some definite outliers in each production system (Figure 1a and 1B). We were able to explain these outliers by maize contents in the respective diet that deviated strongly from the average maize content of the production system (Table 1). Even the unlikely high value in the year-round grazing system could be explained by short-term diet changes (Figure 1A, left): The farm manager of the all-grass farm Q stated that he did not feed any maize in 2013. However, after calling the farm manager again after data analysis and asking for a possible

Table 1. Mean components (% dry matter) of rations fed during housing time for the three production systems as an average over the year.

Year-round grazing	Summer grazing	No grazing
No daily ration	Ration:	Ration:
fodder from pasture	maize: 37%	maize: 41%
+ hay in winter	grass: 37%	grass: 30%
(+ Maize only on farm Q)	concentrates: 25%	concentrates: 29%

explanation of the higher $\delta^{13}C$, he remembered having fed maize silage from a neighboring farm for some time during winter 2013.

Conclusions

$\delta^{13}C$ in cattle hair can be used to estimate the amount of maize (or other C_4 plants) in a range of cattle production systems for at least one year before hair sampling. The method is quite sensitive to short-term diet changes and can be used to retrospectively check the amount of C_4 plants in cattle diet at certain times. Hence, a continuous evaluation of the feeding intensity of the production system, for example systems that are supposed to use grass during summer only, is possible.

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Relationships between milk trans-fatty acids profile and diet fed to cows in Galician (NW Spain) dairy farms

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Abstract

From a collection of 217 tank cow's milk samples taken in Galician dairy farms with varying feeding regimes, it was investigated the relationships between milk *trans* fatty acids (FA) content, milk composition and diet fed to the lactating cows. Samples were grouped into four clusters based on the content of *trans* C18:1 (*t*6-*t*9, *t*10, *t*11 and *t*12 isomers) and rumenic acid (*c*9*t*11CLA) in the milk fat. Average typical diets for each cluster were identified in terms of the relative importance of grass (fresh and ensiled) and starch (from maize silage and concentrate) in the ration fed to the cows. The herbage-based diets had higher mean values of the *t*11/t10 C18:1 ratio compared with the maize-based diets. In contrast, high-starch diets had higher *t*10 C18:1 and lower milk fat contents compared with pasture and silage diets. It was concluded that pasture-based diets produced the healthiest milk FA profile even compared with the linseed oil supplemented diet.

Keywords: pasture, silage, linseed supplement, milk, fatty acids

Introduction

The predominant *trans* fatty acids (*trans*-FA) in human diet are the *trans*-C18:1 isomers, coming either from industrial (partially hydrogenated vegetable oils, which excessive intake is deemed negative for human health) or natural sources from ruminant products (Krettek *et al.*, 2008). Whilst the predominant industrial *trans*-FAs are isomers of *t*9 and *t*10 C18:1, TFAs from ruminants consist mainly of vaccenic (*t*11 C18:1) and rumenic acids (*c*9,*t*11 CLA) that are associated with health benefits (Field *et al.*, 2009). Since fat content and FA composition of dairy milk are mainly dependent on the cows' diet (Elgersma 2015), a study was carried out with the aim of gaining insight in the relationships between diets fed to the cows in commercial dairy farms and *trans*-FA profile of milk.

Materials and methods

This study was carried out using survey data and feed and milk sampling in dairy farms from Galicia (Atlantic-humid NW Spain). Farms (n=45) were selected to represent the broad range of intensification existing in the dairy production system of the region, including both extensive (grazing-based) and intensive (maize-silage based) feeding regimes. Feed components of the ration consumed by lactating dairy cows and bulk tank milk samples were collected five times between October 2013 and September 2014. During the visits, the farmers were asked to describe the lactating cows' feeding as well as milk production, herd number and management. Diet composition was expressed in terms of percentage of each component of the ration in the total dry matter (DM) intake by cow. When the ration included grazed herbage, pasture intake was estimated as the difference between the potential DM intake (DMI) and the total DM of the ration consumed in the barn. DMI (kg d^{-1}) was estimated following the expression $DMI=0.372 \times Yc +12$, where Yc is the average milk yield per cow (L d^{-1}) corrected to the 4% fat. This equation was adapted from NRC (2001) considering cows with 620 kg in the 21th week of lactation. Chemical composition of feed and tank milk samples were estimated in the LIGAL laboratory, respectively, using NIRS (Foss NIRSystems 5000) and FTMIR (Foss Milkoscan FT 6000) calibrations.

FA composition was determined by gas chromatography (GC-FID) following the standards ISO°14156/IDF 172 and ISO°15885/IDF 184. On a final data set of 217 valid observations, a cluster analysis on *trans* C18:1 values and *c9t*11 CLA (expressed in % of total FA) was performed (Proc Cluster and Proc Tree of SAS), followed by an ANOVA analysis (Proc GLM of SAS) for testing the differences between clusters in diet composition and milk physical components and FA profile, after the transformation of the percentage data by means of the arcsine function.

Results and discussion

As can be seen in Table 1, diet composition was different among clusters (P<0.0001) except for the dry forage component. In terms of the importance of grass (fresh and ensiled) and starch (from maize silage and concentrate) in the ration, expressed in %DMI, the four typical diets of clusters 1 to 4 were respectively characterized as high grass-low starch (HGLS: 60.0 and 10.7%), medium grass-medium

Table 1. Diet composition and milk fatty acids composition by cluster.[1]

n	Cluster				*P*
	MGMS	LGHS	HGLS	LGHSL	
	108	37	39	33	
Herd size and dairy cow's yield					
Number of dairy cows	33.4^{b}	51.1^{a}	33.3^{b}	44.5^{a}	<0.0001
Milk yield (corrected 4% fat) kg cow^{-1} d^{-1}	27.7^{b}	32.1^{a}	23.9^{c}	31.9^{a}	<0.0001
Diet composition (% of each ingredient in total DM intake)					
Fresh herbage	12.6^{b}	2.8^{c}	27.4^{a}	9.2^{bc}	<0.0001
Maize silage	18.2^{b}	31.1^{a}	4.0^{c}	34.3^{a}	<0.0001
Grass silage	29.8^{a}	19.3^{b}	32.6^{a}	15.2^{b}	<0.0001
Dry forage	8.5	6.6	7.2	4.0	0.294
Linseed oil	0.0^{b}	0.1^{b}	0.0^{b}	1.0^{a}	<0.0001
Concentrate	30.9^{b}	40.0^{a}	28.7^{b}	36.3^{a}	<0.0001
Milk fatty acids composition (% total FA unless stated otherwise)					
Saturated (SFA)	69.92^{a}	68.62^{b}	66.51^{c}	64.30^{d}	<0.0001
Monounsaturated (MUFA)	25.81^{d}	26.91^{c}	28.35^{b}	30.46^{a}	<0.0001
trans MUFA	1.87^{d}	2.30^{c}	3.17^{b}	3.92^{a}	<0.0001
t6-t9 C18:1	0.43^{d}	0.64^{b}	0.55^{c}	0.84^{a}	<0.0001
t10 C18:1	0.27^{c}	0.57^{b}	0.32^{c}	0.92^{a}	<0.0001
t11 C18:1	1.04^{c}	0.84^{d}	2.09^{a}	1.73^{b}	<0.0001
t12 C18:1	0.14^{d}	0.25^{b}	0.21^{c}	0.43^{a}	<0.0001
t11/*t10* C18:1 ratio	4.35^{b}	1.57^{c}	7.45^{a}	2.24^{c}	<0.0001
Polyunsaturated (PUFA)	3.58^{d}	3.85^{c}	4.40^{b}	4.65^{a}	<0.0001
Linoleic	1.82^{b}	2.25^{a}	1.81^{b}	2.30^{a}	<0.0001
c9-t11 CLA	0.62^{b}	0.54^{b}	1.15^{a}	1.00^{a}	<0.0001
Alpha-Linolenic	0.43^{b}	0.38^{b}	0.69^{a}	0.66^{a}	<0.0001
Omega-6/Omega-3 ratio	3.52^{b}	4.23^{a}	2.48^{c}	3.21^{b}	<0.0001
Milk composition					
Fat (%)	3.89^{a}	3.69^{b}	3.74^{b}	3.47^{c}	<0.0001
Protein (%)	3.23	3.23	3.20	3.23	0.664
Lactose (%)	4.70^{b}	4.76^{a}	4.67^{b}	4.79^{a}	<0.0001

[1] n = number of observations; HGLS = high grass-low starch; MGMS: medium grass-medium starch; LGHS: low grass-high starch; LGHSL: low grass-high starch supplemented with extruded linseed; Figures affected by different letters in the same row are significantly different.

starch (MGMS: 42.4 and 15.7%), low grass-high starch (LGHS: 22.1 and 22.5%) and low grass-high starch, linseed supplemented (LGHSL: 24.4 and 22.2%, with 1.0% of linseed oil). As expected, FA profile was markedly different between clusters. Based on the presence of *c*9-*t*11 CLA and alpha-linolenic acid in milk, both HGLS and LGHSL diets showed the healthiest FA profile and, whilst high grass diet showed the lowest value of the Omega-6/Omega-3 ratio, the linseed supplemented diet had the lowest saturated FA proportion, all differences being significant (P<0.0001). Milk could be classified, from the point of view of human health, as follows: HGLS > LGHSL > MGMS > LGHS, in line with the observation of various researchers (e.g. Elgersma, 2015) indicating that milk from grazing-based systems and from linseed supplemented diets has more n-3 polyunsaturated FA and *c*9,*t*11 CLA, which is considered beneficial for health. Relative percentages of *t*11 and *t*10 C18:1 (expressed on the total *trans* C18:1) were, respectively 66.0 and 10.0% in HGLS, 55.4 and 14.4% in MGMS, 44.3 and 23.4% in LGHSL and 36.7 and 24.6% in LGHS diets, indicating that *t*11/*t*10 C18:1 ratio increases almost linearly with the proportion of fresh and ensiled grass in the ration and decreases in the same fashion with the starch content of the diet. HGLS and LGHSL diets showed the highest values of *t*11 C18:1, followed by MGMS and LGHS, with mean values of 2.09, 1.73, 1.04 and 0.84% total FA, respectively (all values different at P<0.0001). In contrast, the highest values of the isomer *t*10 C18:1 were observed in the two high-starch diets, with mean values of 0.92, 0.57, 0.32 and 0.27% total FA for, respectively, LGHSL, LGHS, HGLS and MGMS diets. An inverse lineal relationship was observed between milk fat content and *t*10 C18:1 content (r=-0.60, P<0.0001), corresponding the lowest milk fat value (3.47%) to the linseed-supplemented, high-starch diet. Results are in accordance with the findings indicating that feeding a grass-based diet enhances the levels of *t*11-C18:1 and *c*9,*t*11-CLA, while feeding cereal-based diets supplemented with oil increases *t*10-C18:1 (e.g. Chilliard and Ferlay, 2004) and this fact is frequently associated with a depression of milk-fat content (e.g. Griinari *et al.*, 1998).

Conclusions

High maize silage-high concentrate diets, supplemented with extruded linseed showed the highest *t*10 C18:1 milk concentrations and the lowest milk fat values. High grass-low concentrate diets showed the highest *t*11 C18:1 milk concentrations. Milk produced in extensive systems, based on grass, showed a healthier FA profile, compared with high starch diets, even when these are supplemented with extruded linseed.

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Key-points for successful pasture-based lamb production observed in Western Sweden

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Abstract

The aim was to observe grazing management and breeding of recruitment sheep by successful lamb producers, and to find ways to improve management by exchange of knowledge and experiences between farmers, advisors and researchers. Eight lamb producers from Western Sweden were included in the observational study that lasted from autumn 2012 to summer 2014. Detailed registrations including pasture nutritional composition, management and animal performance were made on farm level and key-points to successful pasture-based lamb production were summarized. Average herd lamb live weight gain (LWG) from birth to 110 days of age varied from 190 to 380 g day^{-1}. Pasture management, breed and flock size varied and seem to have affected LWG of the lambs. Also, the body condition score of the replacement ewes at mating seemed to influence the LWG of the lambs from birth to 110 days of age (R^2 = 0.36). Lamb producers who set targets and had a strategy with a well-designed work plan met their goals even under varying weather conditions. Regular weighing of lambs is necessary in order to follow up on goals. Key-points for successful pasture-based lamb production, from a specific farm with exceptionally good results, are presented and discussed.

Keywords: grazing, management, replacements, sheep

Introduction

Pasture-based lamb production is the dominant lamb production system both in Sweden and world-wide. There are, however, many different ways of raising lambs on pasture. In Sweden, most farmers let their sheep graze on both semi-natural pastures and on leys from early spring until late autumn. The growth rate of lambs in pasture-based lamb production varies depending on numerous variables. A too low daily live weight gain (LWG) leads to increased costs of production and may lead to lower slaughter income, but also the future production of the replacement ewe lambs are affected by the LWG until mating. One practical measure used in Europe is the weight of the ewe lamb at first mating, which is recommended to be 60% of her adult weight (HCC, 2010). In this on-farm study, which included eight farms with pasture-based lamb production with a range of differences in genetic and environmental differences, the aim was to find key-points in the production used by successful lamb producers. The data was collected by on-farm observations and included planning and execution of plans in the areas of grazing management and replacement-ewe management.

Materials and methods

Eight farms were included in the study that lasted from autumn 2012 to summer 2014. The project noted what the producers had aimed for in their production and what they thought they could improve. Also, data were collected giving background information from all farms. In addition to individual farm visits, the whole group gathered on farmers' farms to exchange experiences and plan activities and to discuss the results that came up during the project. The ewes were weighed and body condition was assessed before mating, after lambing and at weaning, according to Thomson and Meyer (1994). The lambs were weighed at birth, at turnout and at weaning as well as continuously the weeks before slaughter. The replacement ewes were followed up until they lambed for the first time. During the grazing period, the pasture height was measured once per month in the field from where the sheep were turned out. Samples were cut in the same field for analysis of nutrient and mineral contents. Farmers kept grazing diaries with details of grazing type, field area, sward type etc. Lambing took place in March to May depending on breed, and ewes and lambs on all farms were turned out on pasture as soon as possible. A grazing plan was made on each farm where farmers set their goals for their grazing strategy for the summer. On most of the farms where the farmers reached their targets, they worked actively with grazing management including rotational grazing, trimming of paddocks, supplementary feeding when necessary and allowing different categories of animals to graze on different pastures (grouping) etc. Also, the most successful farmers did not suffer from any parasitic problems, as the faeces were analysed and the sheep were treated based on parasitic species. Results from one of the most successful farmers are presented and discussed.

Results and discussion

One of the most successful farmers (No. 3) reached an average of 1.8 weaned lambs per replacement ewe (n=22) put to the ram. All the older ewes were also pregnant and had an average of 2.3 weaned lambs per ewe (n=70). All lambs were either sold, slaughtered or pregnant when the grazing period ended. Also, the ewe lambs from this farm reached, but did not exceed, the optimum BCS of the ewes at mating (Figure 1). Furthermore, the average BW was 63% of the adult weight at mating and the sheep grazed only leys (Figure 1). The first ley grazed had not been used for sheep the year before and the replacement ewes were grouped and kept in one group after weaning until next year's grazing period started. The weather varied between years, with 2013 being generally warmer and drier than 2012 in this part of Sweden. Averaged over farms, the protein content of the grass seemed to be linked to the LWG of the lambs, and the protein content of semi-natural pastures declined more rapidly than that of cropped leys during the summer.

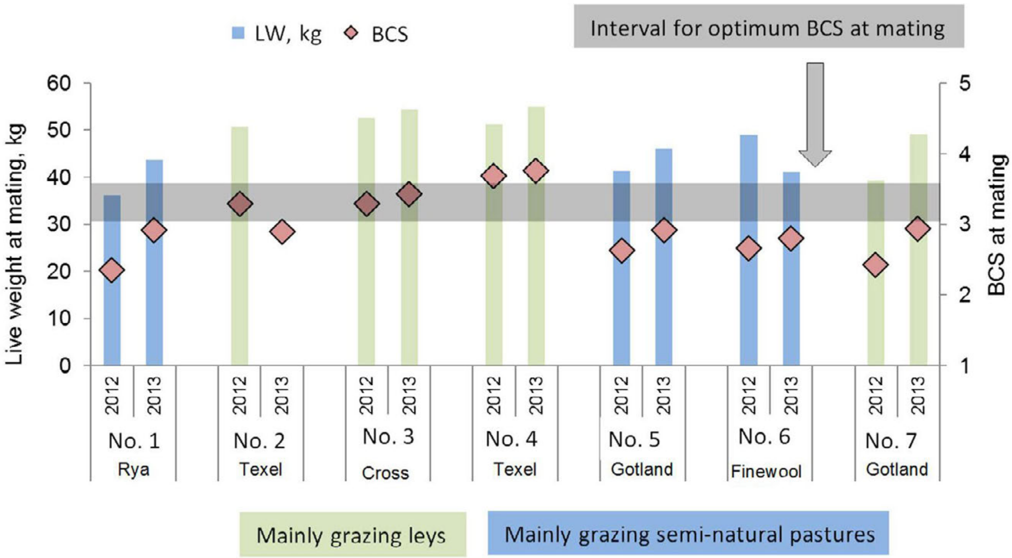

Figure. 1. Live weight and BCS of replacement ewes at mating in 2012 and 2013.

Identified key-points for successful pasture-based lamb production:

- Weighing and condition scoring of ewes at mating, at end of grazing period and at start of grazing period.
- Strategic grouping when indoors, both during pregnancy and lactation.
- Feeding grass/clover silage with high feed value supplemented with pelleted concentrate.
- Parasitic control through analyse and treatment of specific species.

- Grazing plan with rotational grazing, trimming of pastures and supplementary feeding when needed.
- Adjusted grazing area by grouping and mobile fences.
- Regular weighing of lambs on pasture.
- Active selection of replacement ewes meeting criteria.
- Keeping replacement ewes in a separate group from weaning until next grazing season.
- Using cross-bred ewes from fertile breeds (e.g. Fine wool/Dorset) mated with a meat-breed (e.g. Texel).

Conclusions

Lamb producers who set targets and build strategies with a well-designed work plan generally meet their goals quite well even under varying weather conditions. The key-points presented in this paper highlight the importance of weighing, condition scoring, grouping, silage quality, parasite control, grazing management and active selection of replacement ewes. As the protein content of the pasture seemed to be linked to the LWG of the lambs, semi-natural pastures are most suitable for use early in the spring, as the content of protein and metabolizable energy declines more rapidly on these pastures than on leys.

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Theme 2.
Forage and pasture quality evaluation

Forage quality evaluation – current trends and future prospects

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Abstract

In many counties worldwide, ensiling is the major form of forage conservation. However, dry matter (DM) intake (DMI) by ruminants is often lower for silages than for the herbage in fresh or hay form. The pattern of fermentation products as a result of the ensiling process is seen as the main factor being responsible for reduced DMI. As the nutritive value is very variable evaluating forage quality correctly with appropriate methods is indispensable. Fermentation related factors are indicative of the efficiency of the conservation and of the modifications of carbohydrate and nitrogen fractions during the preservation; however, their effect on DMI is still often unknown. Reductions in intake of silage have been related to acidity due the presence of free acids and the high proportion of total acids as lactic acid, to poorly fermented silages and to acetic acid as single fermentation product. The potential DMI is maximized with restricted fermentation in low DM, early harvested grass silages. Losses occurring during silage making and utilisation can exceed 200 g kg^{-1} of DM and need to be considered for an appropriate evaluation of silage production and utilization.

Keywords: fermentation, forage, intake, silage

Introduction

Although fresh, unpreserved herbage is the major form of forage from grassland in many countries worldwide and in some European areas, too, the geographic location and extended north-south gradient of Europe often require that the majority of harvested grassland biomass is preserved as hay or silage. In Nordic countries and regions with maritime climate, little hay is produced and the vast majority of harvested biomass is preserved as silage. Silage making is less weather-dependent than hay making, but the production still demands good management to prevent excessive losses and to minimize reductions in nutritive value of the harvested crop. The question arises whether type of preservation has an impact on the choice of methods applied to evaluate forage quality. To address this question, this paper focuses on similarities and dissimilarities of silage compared to fresh herbage and hay relative to forage quality evaluation. Aspects of protein value and N compounds will not be covered as these are addressed by others in this volume.

Brief review of forage quality determinants and impacts

Ensiled forage offered to ruminants often results in a lower voluntary dry matter (DM) intake (DMI) as compared to the corresponding fresh (Donaldson and Edwards, 1976) or dried feed (Thiago *et al.*, 1992). Intake may be decreased compared to hay and fresh forage by more than half (Campling, 1966). However, silage is important for productive and efficient ruminant livestock farms, especially in humid and temperate areas, where DM and quality losses in making hay may be excessive due to wet weather between cutting and harvesting the crop (Pahlow *et al.*, 2003). In general, ensiling is less weather dependent than hay production due to the shorter period of time between cutting and harvesting. A disadvantage of both types of forage conservation is loss of feed value compared to that of the original crop. The extent of the loss depends on the crop management, resulting in large variation in nutritional value and fermentation quality (McDonald *et al.*, 1991). The quality of silage correlates with the pattern of fermentation, which may be the primary cause for decreased DMI in ruminants offered silage-based diets (Eisner *et al.*, 2006).

Crop related factors of forage quality are primarily related to maturity stage at harvest. Digestibility of organic matter (OM) is probably more representative of the maturity stage at harvest than chemical entities such as cell-wall (neutral detergent fibre, NDF) or crude protein. Lignification of the cell wall fraction increases with plant maturity and reduces digestibility. Lower temperatures, long day, and high solar radiation during the growing season at northern latitudes promote DM accumulation, but not lignification (Van Soest *et al.*, 1978). Therefore, digestibility can be maintained at high levels (700-750 g kg^{-1}) even at high NDF content (600 g kg^{-1} DM) at northern latitudes (Huhtanen, 1998). The strong environmental effect on fibre content and quality favours the approach to estimate digestibility or energy value of forage for specific geographic origin or forage types. Krizsan *et al.* (2012) followed the latter approach to compare different *in vitro* and in situ methods applied to empirical and mechanistic predictions of *in vivo* OM digestibility (OMD). Compared with the *in vitro* laboratory methods, indigestible NDF (iNDF) used in forage-specific equations improved overall predictions of forage *in vivo* OMD. Others have derived generalized, empirical equations to estimate energy values for ruminants of fresh or preserved grassland forage. Based on a comparison of specific and generalized equations for 506 grass products (fresh, silage, hay), GfE (2008) in Germany concluded that 'prediction equations for grass products that include an *in vitro* variable do not need to differentiate between products and cut numbers.'

To get a more complete assessment of silage quality, fermentation variables are beneficial not only in routine analyses. Fermentation related factors are indicative of the efficiency of the conservation and of the modifications of carbohydrate and nitrogen fractions during preservation. Extent of fermentation is strongly correlated to the DM content of the silage (Muck, 1990). Based on the digestible OM in the DM, and fermentation characteristics, the silage is given an index, relative to that of a well-fermented silage of high digestibility with a silage DM intake (SDMI) of 100% (Huhtanen *et al.*, 2002). A modified form of the SDMI index developed by Huhtanen *et al.* (2002) has been applied in Norway to estimate the relative intake potential of silages (Flittie Anderssen, 2003). To assess fermentation quality, concentrations of ethanol, individual organic acids and the pH may be determined. Besides ethanol and organic acids, a large number of mainly unknown components in grass silages may occur. Mo *et al.* (2001) reported fifty-one identified fermentation products in collected field samples of grass silage. The relevance of these components on silage quality is still largely unknown.

Current trends in forage quality determination

It is generally assumed that the intake potential of herbage is reduced as a result of ensiling. Mayne and Cushnahan (1995) reviewed literature data from thirteen studies summarising 432 individual observations and concluded that DMI was on average reduced by 27% when comparing cut grass with silage. The extent of intake variation varied widely between trials and the largest range in reduction of silage intake was from 1 to 64% (Demarquilly, 1973). The wide variation in reported reductions in SDMI was explained by the lack of correction of DM content for losses of volatile components during oven drying (Mayne and Cushnahan, 1995). Further, no adverse effects on intake were observed when restrictively or extensively fermented silage, both well fermented, was compared with fresh herbage.

Grazed grass versus silage

Mayne and Cushnahan (1995) further pointed out that lower digestibility of herbage at ensiling relative to that of grazed material could explain most of the variation in intake observed in practical farming situations. Common farm practise is shorter regrowth intervals with grazed grass compared to herbage cut for ensiling. Increasing number of cuttings to improve silage digestibility will decrease DM yields and increase total cost of silage production, but generally promote a higher intake. However, when harvesting is by the animal rather than a machine there will be no direct financial penalty by grazing swards with low herbage mass, but there will still be conflicts between yield, quality, intake, and animal performance (Wilkins, 2003). The advantages with grazed grass compared with silage feeding, except lower costs of DM, seems primarily to be a higher feeding value and(or) more efficient nutrient utilisation, and generally maintained animal performance with lower inputs of concentrate feed (Wilkins, 2003).

Hay versus silage

Reductions in intake observed with cattle offered grass silage compared with hay prepared from the same sward have been attributed to the end products of fermentation (Gill *et al.*, 1988). Gill *et al.* (1988) expressed that a more complete understanding of the factors controlling the size of each meal, should lead to a more comprehensive understanding of the major factor limiting daily intake. Experimental designs should allow measuring effects of the gas phase on physical distension (Gill *et al.*, 1988). Distension signals are, except for digestion kinetics, also related to specific gravity, volume, and rate of particle size reduction and passage of the feed (Mertens, 1994). Gill *et al.* (1988) concluded that intake of silage responds to some products released from the feed, or a rapid increase of ruminal VFA concentration, in addition to distension signals without ranking the importance of these factors with regard to daily intake. Shingfield *et al.* (2002) compared silage with hay harvested one week later, but from the same field, and when correcting DMI for decreases due to lower digestible OM in the DM, intake data of treated silages was comparable with that of hay. Dulphy and Van Os (1996) reviewed the DMI by dairy cows fed silage of good quality compared with hay prepared from the same parent crop, and reported a 6% increase for eight comparisons and a 5.5% decrease for three comparisons in the literature. As with feeding cut or grazed grass, it was concluded that when silage is well fermented no adverse effects on DMI can be expected (Dulphy and Van Os, 1996). When intake of silages was compared with the intake of hay the variation was from 26 percentage units reduction to 9 percentage units increase, and the silage giving highest intake showed restricted fermentation combined with second highest content of water-soluble carbohydrates (Krizsan and Randby, 2007). The silages displaying reductions in intake of silage DM compared with hay DM, showed signs of poor preservation, or were those with highest content of lactic acid (LA; Krizsan and Randby, 2007).

Silage acidity and intake

Low intake of silage has been related to acidity due to the presence of free acids and the high proportion of total acids as LA. Thomas *et al.* (1980) and Choung and Chamberlain (1993) observed lowered DMI by growing cattle and lactating cows, respectively, with increases in silage LA content. The addition of LA to silage produced a linear increase in the molar proportion of propionate in ruminal fluid (Choung and Chamberlain, 1993). Thomas and Wilkinson (1975) observed a more pronounced effect on blood pH and acid-base balance of the animal than on ruminal pH by neutralization of silage acids with sodium bicarbonate. This suggests that the rapid ruminal metabolism of LA to propionate would influence the absorption of volatile fatty acids (VFA), which is in accordance with the observed tendency of increased plasma insulin concentrations (Choung and Chamberlain, 1993).

Intake variation due to fermentation quality

Traditionally reductions in intake of silage induced by fermentation end products have been evaluated in comparisons with grass or hay of equal maturity. It has, however, been observed that silage intake can be higher than that of hay (Dulphy and Van Os, 1996; Krizsan and Randby, 2007). This implies that a more accurate estimate of variations in SDMI due to fermentation quality would come from comparisons with a high-intake silage rather than of hay. The estimated variation in SDMI when intakes for all silages were compared with the silage giving highest intake indicated reductions between 6 and 32%, with an average reduction of 13% (Krizsan and Randby, 2007). Several fermentation products have been claimed to be responsible of the reductions in intake observed on silage diets. Naturally, the effect of other acids, not only LA, in silage has been examined regarding their impact on intake. An effect of VFA on intake seems to be more likely when they are provided in combination rather than individually (e.g. Gill *et al.*, 1988; Faverdin *et al.*, 1992; Anil *et al.*, 1993).

Krizsan *et al.* (2007) suggested that extent and type of silage fermentation influenced intake and that the potential intake of silage DM is maximised with restricted fermentation in low DM silages. Keady and Murphy (1997) observed higher intake by lactating cows given grass silage treated with formic acid at low and high levels than when fed silage untreated or treated with a bacterial inoculant. The increase in intake with formic acid treatments was related to improvements in silage fermentation quality indicated by lower concentrations of VFA and NH_3-N. However, the LA content of silages treated with low level of formic acid and bacterial inoculant was comparable (Keady and Murphy, 1997). Moreover, Keady and Murphy (1998) observed a clear preference of the restrictively fermented silage treated with high level of formic acid when a choice between the silages was allowed. Mayne and Cushnahan (1995), Selmer-Olsen and Mo (1997), Huhtanen *et al.* (1997), and Shingfield *et al.* (2002) observed improved intake by lactating cows of 0.8, 1.0, 1.5, and 0.4 kg day^{-1} of DM, respectively, with silages of restricted fermentation when compared with one displaying extensive fermentation. The improved intake of restrictively fermented silage was reflected in improvements of total DMI (Huhtanen *et al.*, 1997; Shingfield *et al.*, 2002). However, the differences in SDMI were not significant (Mayne and Cushnahan, 1995) or only tended to be significant (Huhtanen *et al.*, 1997; Shingfield *et al.*, 2002). Selmer-Olsen and Mo (1997) reported a significant increase (P<0.01) in mean SDMI of restrictively fermented silage when compared with silage of extensive fermentation.

Steen *et al.* (1998) put the relevance of including fermentation variables in prediction equations of SDMI in perspective by modelling intake of grass silages displaying wide ranges in crop- and fermentation-related variables. Significant relationships were reported between intake and fermentation products but with only small R^2 values (0.04 to 0.11; Steen *et al.*, 1998). The most important factors influencing SDMI were related to protein and fibre fractions, and *in vivo* DM and OM digestibility. The curvilinear relationship between DM content and intake indicated a maximum intake at a DM concentration of 320 g kg^{-1} even though the linear relationship was a significantly better fit (Steen *et al.*, 1998). Rook and Gill (1990) found that intake did not increase already at DM concentrations above 250 g kg^{-1}.

Losses occurring at different scales

Losses occurring during forage preservation along the whole silage production chain have only recently gained more thoughtfulness, not only on a laboratory scale but including farm scale estimates. Quantification of total losses has long been neglected and both inevitable and evitable losses have not been accounted for. More than 25 years ago, McGechan (1990) evaluated data on hay and silage storage losses, and developed models for predicting storage losses, Compared with hay, losses occurring during the storage of silage are greater, typically 20% or more, yet the subject is complex. Material which is lost upon air infiltration is invisible, and fermentation losses caused by malfermentation are entirely from rapidly degradable energy components of the silage. Control of DM losses (DML) is a major concern of forage conservation systems, yet determination of DML during hay and silage storage is difficult and time-consuming. The lack of a practical way of measuring DML has contributed to the poor adoption of good practices. The availability of a practical, easy, and economic technique capable of estimating on-farm DML would facilitate extension work. Jaurena (2012) used a Monte Carlo simulation approach to quantify DML during forage conservation and concluded that there is a reasonable basis to predict DML by compositional changes. Köhler *et al.* (2013) measured losses during conservation of maize, grass and lucerne in farm-scale bunker silos, from field to feeding. With well-fermented silages, 80 g kg^{-1} DM losses occurred but much higher values of up to 260 g kg^{-1} were observed with mal-fermented or aerobically spoiled silages. With the latter, not only DM losses will occur but also reduced feeding value and DMI intake. Gerlach *et al.* (2014) observed a reduction in DMI and preference of well-fermented grass silages by goats at the sixth and eighth day of aerobic exposure, also at apparently low levels of deterioration. This underlines that unidentified, volatile compounds might affect preference and DMI. Goats (and other ruminants) may detect subtle differences caused by oxygen ingress, sometimes even before an increase in temperature or changes in chemical composition occur.

Relationships between dry matter content, fermentation and intake

Mo *et al.* (2001) pointed out propanol and dimethyl sulfide as components negatively related to intake of silage. Randby (2006) observed that reductions in SDMI equalled the amount of added propanol when silage-fed lactating cows were added 200 g propanol or 200 g propanol + 4 g dimethyl sulphide and concluded that total DMI was not affected by treatment. Most attempts to isolate specific fermentation products involved in controlling silage intake have failed to conclusively pinpoint any particular product as responsible for reduced silage intake.

To further elucidate the relationship between fermentation products and DMI over a wide range of silage DM concentrations, Eisner (2007) conducted a meta-analysis to quantify effects of fermentation products on DMI. The analysis included 70 trials with 321 observations for diets when forage and concentrate were offered separately and 15 trials with 63 observations for total mixed rations. The concentration of acetic acid was closely and negatively correlated with SDMI when concentrate and forage were offered separately (Figure 1).

The effects of other VFA and of LA were indistinct. Total acid concentration was closely and negatively related to total DMI in diets fed as total mixed rations. Current equations for predicting feed intake and performance of dairy cows need further development and refinement (Eisner, 2007). Recently, Weiss and Auerbach (2013) studied the incidence of volatile organic compounds, including ethyl esters of LA and acetic acid, in grass silages. Based on all available data (n=1,148) from different types of silages, a generally valid model was proposed to predict total ester concentrations (y) from ethanol content (x): $y = 114x$ (R^2=0.76).

Mechanisms behind reduced intake of silage including metabolic and production effects on the animal would be a relevant point but this is outside the scope of the current article.

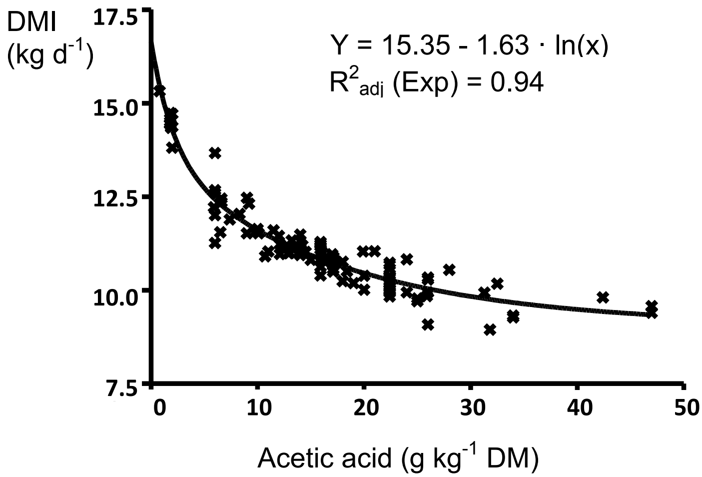

Figure 1. Effect of acetic acid content on dry matter (DM) intake (DMI) of wilted silage by dairy cows (Eisner, 2007).

Perspectives and conclusions

Chemical analysis of the feed in order to identify all related substances should be combined with investigations of physiological responses by the animal at the time of feed intake. For the future it would be desirable to include more silage compounds in routine forage analysis to obtain more data about possible relationships among specific compounds. To elaborate more profound causalities the respective features need to be better characterized and new features should be created. Studies involving integration of physical and physiological regulation would be an important step forward, for example the possible impacts of silage chop length on fermentation and physiological processes. Silage production will remain a key part in ruminant production systems across Europe. This demands continuing approaches for production of high quality silages involving improvements in management practise to minimise losses and maximise the preservation of the inherent feeding value of the parent crop.

At low DM concentrations, early harvested grass silages displaying restricted fermentation seem to be a preferable quality, promoting a possible increase in DMI, even though the experimental evidence is ambiguous, but more clearly inducing a positive production response and increased feed efficiency by lactating cows. Little production responses will occur from wilting when the ensiled material is treated with formic acid. Wilting mainly seems to induce positive responses on DMI of early harvested material treated with inoculants or untreated material. Generally, nutrient recovery and quality is good when fermentation has been restricted with acid-based additives in low DM grass silages. To further minimise the risk of clostridial fermentation, low DM grass should preferentially be wilted to a DM content of 300 g kg^{-1}. This is also the level of DM content often recommended giving smallest total losses, i.e. in the field, from fermentation, and from effluent. The importance of quick attainment and maintenance of anaerobic conditions in the silo to limit proteolytic activity and unwanted microbial growth is inevitable. Changes in chemical composition during ensilage impact the energy supply to ruminal microbes. Paralleled with the increase in degradable N compounds it will affect the balance of absorbed nutrients. Adjusting the ratio of degradable protein and fermentable energy supply in the rumen has been suggested to be the best opportunity when trying to improve the efficiency of N use.

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Sensing grassland quantity and quality – new technologies in the field and laboratory

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Abstract

Grassland systems frequently exhibit small-scale botanical and structural heterogeneity with pronounced spatio-temporal dynamics. Such features pose particular challenges for any sensor application. Regarding these problems and the considerable costs and frequently limited spatial resolutions of many available remote sensing (RS) systems, it is comprehensible that up to date hardly any commercial application of RS for practical grassland farming exists in Europe. However, considering the developments in sensor performance, data processing and analysis and modelling over the recent decades, this paper identifies significant advances in RS for grassland research and practice and reviews the most important sensor types and corresponding findings in research. Beside improvements of single sensor types, the development of systems with complementary sensors are seen as a most promising research area, which will help to overcome the limitations of single sensors and to provide better information about grassland composition, yield and quality. From an agronomic point of view, thematic maps from farm fields are suggested as the central outcome of RS and data analysis, which represent the relevant grassland features and constitute the basis for various farm management measures on the strategic, tactical and operational level. The overarching goal will be to generate cheap, appropriate and timely information and to provide it to farmers to support their decision-making.

Keywords: remote sensing, spectrometry, radar, ultrasound, grassland yield, forage quality

Introduction

Traditional techniques based on field measurement (e.g. by weighing) are the most accurate ways for collecting biomass data. However, these approaches are often time consuming, labour intensive, and difficult to implement, especially in remote areas; also, they hardly can provide the spatial distribution of any biomass parameter in large areas. This is where remote sensing (RS) comes in, with its repetitive data collection and digital format. RS allows a fast recording and processing of large quantities of data and making it the primary source for large area biomass estimation. RS has been defined as 'the field of study associated with extracting information about an object without coming into physical contact with it' (Schott, 2007). Within the context regarded, remote sensors are used to capture specific information or sample or canopy characteristics without the need to directly measure the particular information of interest, but simply provide data from which the desired information can be extracted based on some observables of the remotely viewed sample or vegetation.

RS systems provide different information features, such as in spectral, radiometric, spatial, and temporal resolutions, and in polarization and angularity (Barnsley, 1999). There are many sensors available with different characteristics of spectral, spatial, and temporal resolutions used for biomass and quality estimation limited by availability, efficiency and costs. Recognizing and understanding the strengths and weaknesses of different sensor types is essential for selecting suitable sensor data for biomass estimation. Optical remote sensing, radar and light detection and ranging (LiDAR) sensors provide the three main sources of remotely sensed data for biomass and quality estimation, but ultrasonic sensors may also provide an interesting potential.

This review focuses on RS in grasslands under moderate climate conditions, it barely covers the vast literature on RS in rangeland ecosystems (including savannas) in the tropics and sub-tropics, as the results can hardly be transferred to the (among other things usually more intensive) grassland systems in moderate climates. In remotely sensed data, the task of raw data pre-processing is essential to ensure a high quality dataset containing the desired information of surface reflection. There exists a wide range of methods to overcome the problem of signal noise caused by atmospheric conditions or backscatter effects of the signal (e.g. McGovern *et al.*, 2002; Canty *et al.*, 2004; Franklin *et al.*, 1994; Hadjimitsis *et al.*, 2004; Ricketts *et al.*, 1992). The issues of data correction will not be further covered in this review.

Sensors: technical principles and recent research findings

Photography

Photography creates durable images by recording light or other electromagnetic radiation, either electronically by means of an image sensor, or chemically by means of a light-sensitive material such as photographic film. Film usually records radiation over a wavelength range similar to the human eye (0.3 to 0.9 mm versus 0.4 to 0.7 mm). Given appropriate ground reference data, accurate measurements of positions, distances, directions, heights, volumes, areas and slopes can be obtained from photograph. Photogrammetry is the science of making measurements from photographs to determine the condition, shape and position of the given objects of interest. Nowadays, digital photography is the most used form of photography in remote sensing, which uses cameras containing arrays of electronic photodetectors to capture images focused by a lens. The most common detector in a digital camera (charge-coupled device image sensor) provides data in the blue, green and red area of the visible spectrum. Digital image analysis (DIA) have been successfully applied to agriculture to identify and estimate biomass and locate individual plants. DIA can be used to distinguish between crops and weed species (Hague *et al.*, 2006; Onyango *et al.*, 2005). A canopy of diverse grassland plants presents several difficulties to DIA, including the diversity of optical plant properties within a mixed sward, varied leaf colours and shapes, overlapping of leaves and tillers, shadows on leaves and soil, non-uniform soil background, and different leaf appearances during the growing season. *Rumex obtusifolius* L. was detected in mixed grassland swards by recording images with a remote-controlled vehicle in the field, segmenting the images using homogeneity threshold and defining objects and features describing shape, colour, and texture (Gebhardt *et al.*, 2006; Gebhardt and Kühbauch, 2007). Based on results from a pot experiment digital image analysis (DIA) was suggested to assess the legume contribution in legume-grass mixtures (Himstedt *et al.*, 2009). A revised model was validated with weekly sampled data from spring, summer and autumn cuts of field-grown swards of red clover- and white clover-grass mixtures (Himstedt *et al.*, 2010), and a high prediction accuracy (r^2=0.98, SE = 6% of DM) was obtained across a wide gradient of growth stages. However, information from photos is restricted to the canopy surface, which may limit the applicability in higher forage crops, like maize or cereals.

Spectroscopy

Spectroscopy is the study of the interaction between energy and matter and the data obtained displays a spectral signature or spectrum for each pixel/point/measured location. Spectroscopic data represents a plot of the response of interest as a function of reflectance value and wavelength. Spectroscopy utilization has undergone considerable change in Europe in the past two decades (Dale, 2014).

Lab ultraviolet-visible (UV/VIS) spectroscopy: Absorption of this relatively high-energy light causes electronic excitation, molecular vibrations and electronic transitions brought about by UV radiation. The easily accessible part of this region, the UV wavelengths region is between 190 and 380 nm, while the VIS wavelengths region is from 380 to 750 nm (Workman, 2000), that absorb only if conjugated pi-electron systems are present (Workman, 2000). With the help of the UV spectroscopy inorganic and organic components of the samples were identified, e.g. crude protein and amino acids from manure samples (Weckhuysen, 2004).In UV spectroscopy, the sample is irradiated with the broad spectrum of the UV radiation, and UV light, which is not absorbed, will pass through the sample and will be recorded. The UV-VIS technique was used in the context of nitrate determination in plant material (Heanes, 1982), ammonia determination in banana by-products for feeding small ruminants (Álvarez *et al.*, 2015) and of methane emission (Bloom *et al.*, 2010).

Lab near infrared (NIR) spectroscopy: Absorption of this lower energy radiation causes vibrational and rotational excitation of groups of atoms within the molecule. Because of their characteristic, absorption identification of functional groups is easily accomplished. The measured spectra are used to determine the chemical and physical composition. The correlation between structure and spectrum provides a base for a cause-and-effect relationship between the spectra (instrument response) and reference data (analysis), with the aim of providing a scientific basis for different multiple choices on infrared spectroscopy (Workman, 2005). The infrared spectral regions of the electromagnetic spectrum extend from 780 to 100,000 nm and are classified into near infrared (NIR), middle infrared (MIR) and far infrared (FIR) (Manley *et al.*, 2008). NIR spectrometry is used commonly to determine forage composition and quality such as protein, dry matter, ash, fibre, fat, neutral detergent fibre, acid detergent fibre, lignin, digestibility and crude energy or the compound feeds for cattle by faeces analysis (De Boever *et al.*, 1995). In recent years, NIR spectrometry has been used for plant selection and plant nutritive value evaluation, silage and crop information, feed ration balance and quality (Decruyenaere *et al.*, 2009). Recently, biomass determination (Laurens *et al.*, 2013) and grassland species discrimination (Dale, 2014; Wachendorf *et al.*, 1999) was accomplished. Whatever its application, this technology is only as good as the calibration data derived from reference analysis. Once adequate calibration models are in place, NIR spectrometry can provide rapid, inexpensive and accurate assessments of feed composition in a large number of samples (Weiss *et al.*, 2007).

Nuclear magnetic resonance (NMR) spectroscopy: Absorption in the low-energy radio-frequency part of the spectrum causes excitation of nuclear spin states. NMR spectrometers are tuned to certain nuclei. The radio frequency of the absorption spectra of atomic nuclei in substances is subjected to magnetic fields (Svanberg, 2012). The spectral dispersion is sensitive to the chemical environment via 'coupling' to the electrons surrounding the nuclei. For a given type of nucleus, high-resolution spectroscopy distinguishes and counts atoms at different locations in the molecule (Svanberg, 2012). NMR is used in different context for different aims: to solve compounds structure like synthetics pesticides (Raikwar, 2013), analysing species, biomass, and vitality of plant roots (Rewald and Meinen, 2013), biomass components (Mori *et al.*, 2015), trends within series of compounds, e.g. protein behaviour of condensed tannins (Zeller *et al.*, 2015) and ligand binding (Santos *et al.*, 2015). Actually, the portable NMR was developed for measuring objects that are too large or too sensitive to be move into the laboratory, such as tree branches or growing fruits on a plant (Windt *et al.*, 2010). NMR could be further used, i.e. for measuring the anatomy and the dynamic changes of tree, plants and fruits, sap flow and water transport in the intact plant (Windt and Blümler, 2015), e.g. the allocation of carbon to various plant organs (Jahnke *et al.*, 2009).

Field spectroscopy: Spectroscopy makes use of electromagnetic radiation that normally ranges from 0.4 to 14 mm wavelength and measures the diffuse reflectance properties of vegetation mostly with passive sensors, which completely rely on the sun's radiation. Healthy green vegetation typically shows a 'peak-and-valley' pattern of spectral reflectance. In the visible spectral region (0.4 to 0.7 mm) valleys occur due to energy absorption by plant pigments both in the blue (chlorophyll b, carotenes) and red (chlorophyll a) bands, resulting in our perception of healthy plants as being green. As plants senesce or become subject to some form of stress, absorption in the blue and red bands is reduced and the plants are perceived as yellow, i.e. a combination of green and red. Dying plants exhibited a brown colour, as leaf reflectance is decreased over the entire visible range. Radiation in the shortwave infrared (1.3 to 3 mm) is essentially absorbed or reflected, depending on the water content and thickness of leaves. While multispectral sensors measure the reflectance in multiple spectral bands, hyperspectral sensors acquire data in several hundred very narrow, contiguous spectral bands throughout the visible and NIR portions of the spectrum. The result is a mathematical model between canopy properties (e.g. yield, protein concentration, species diversity) and the full sensor system response is referred to as spectrometer calibration. Spectral sensors have raised considerable interest as a potential tool for prediction of biomass in pastures. Spectral reflection measurements have been widely used for the characterization of grassland biomass, obtained from hand-held hyperspectral radiometers (Chen *et al.*, 2009; Mutanga and Skidmore, 2004; Vescovo *et al.*, 2012) but may contain large amounts of redundant information. For practical implementation at field scale, the limitation of wavebands as vegetation indices is desirable. Vegetation indices (VIs) are widely used in RS models for estimation of various crop characteristics (Hatfield and Prueger, 2010; Huang *et al.,* 2012) like grassland biomass (Boschetti *et al.*, 2007; Numata *et al.*, 2007; Todd *et al.*, 1998). However, the performance of VIs is highly site and sensor-specific (Huang *et al.*, 2004). Selection of distinctive narrow bands from hyperspectral data, e.g. according to the NDVI-type formula have shown improvements to traditional VIs (Thenkabail *et al.*, 2000; Reddersen *et al.*, 2014). However, difficulties with biomass prediction occurred at advanced developmental stages of grassland vegetation, as the ability of the reflectance sensor to detect canopy characteristics could be limited by the presence of a high fraction of senescent material in biomass (Yang and Guo, 2014). Further limitations may originate from soil background effects (Boschetti *et al.*, 2007), atmospheric conditions (Jackson and Huete, 1991), grazing impact (Duan *et al.*, 2014) and heterogeneous canopy structures due to mixed species composition and a wide range of phenological stages (Biewer *et al.*, 2009a, b). For the assessment of forage quality parameters using proximal sensing of pasture canopy reflectance broadband multispectral sensors are considered to have limitations in providing accurate estimates of vegetation characteristics (Thenkabail, 2012), while hyperspectral sensors with narrow and near-continuous spectra allowed much more detailed spectral information, offering significant improvements over broadband sensors. Partial least square regression (PLSR) is a technique for analysing hyperspectral datasets that employs the whole range of hyperspectral data in the analysis. Several studies have shown that PLSR is a powerful tool to accurately predict forage quality constituents in the field condition (Biewer *et al.*, 2009a; Starks *et al.*, 2004; Li *et al.*, 2014a). However, due to costs and complexity of hyperspectral data, reducing the spectral data range and identification of the best spectral features of hyperspectral information would facilitate simple sensor applications in the field (Biewer *et al.*, 2009b; Li *et al.*, 2014; Reddersen *et al.*, 2014). Comparisons between traditional VIs and hyperspectral narrowband VIs showed a higher accuracy for the latter for various vegetation characteristics (Fricke and Wachendorf, 2013; Thenkabail *et al.*, 2000).

Spectral imaging

Spectral imaging is the combination of two different sensing modes: imaging and either multi- or hyperspectral spectrometry. Hyperspectral imaging sensors are able to simultaneously capture both the spatial and spectral content of remote scenes with high spatial and spectral resolution and coverage. The resulting data product is sometimes called a hypercube, which can be imagined as a three-dimensional dataset, where each two-dimensional pixel contains a whole spectrum (as third dimension) whose signatures are related to the materials contained within it. The size of each pixel depends on the mounting height of the scanner and its field of view, and can vary between the sub-centimetre range (with proximal measuring distance) and several meters (when the sensor is mounted on an airplane or satellite). Remarkably, most studies on remotely sensed data for the estimation of grassland and rangeland biomass were conducted in tropical savannas, since these ecosystems account for 30% of the primary production of all terrestrial vegetation, or in semi-arid to arid rangelands of Asia or North America. Contrary, comparable studies on grasslands in temperate climates are rare (Kumar *et al.*, 2015). Schut *et* *al.* (2006) used a hyperspectral imaging sensor system recording reflexion intensity from 439 to 1,680 nm. When predicting grassland yield on experimental fields they obtained R^2 values of 0.91, 0.86 and 0.96 for Lolium perenne-dominated, heterogeneous and grass-clover swards, respectively, with a root mean square error of 0.34, 0.48 and 0.17 t DM ha^{-1}. However, application of the sensor system in fields from two farms at several dates during the growing season produced larger errors of 1.4 t DM ha^{-1}, with a wide range among single dates. The authors attributed this phenomenon to system instability and environmental disturbances (effects of weather and location). Givens and Deaville (1999) reported that similar problems of method incompatibility when using a near-infrared spectroscopy calibration set by three different consultants. For sugar concentration, relative errors were between 15 and 16% and for crude fibre, neutral detergent fibre, acid detergent fibre, and digestibility, relative errors were between 3 and 5%. Mutanga and Kumar (2007) estimated and mapped grass phosphorus concentration in African rangeland and obtained a R^2 of 0.63 with RMSE of 0.07 for the test dataset.

The increasing ability of remote-sensing technologies to rapidly deliver data on habitat characteristics, like distribution of individual plant species, habitat types and/or communities, across a range of spatial resolutions and temporal frequencies is increasingly sought-after in conservation management (Mairota *et al.*, 2015). Recent studies estimating diversity with RS techniques focused on mapping species distribution and alpha diversity (Hall *et al.*, 2010; Psomas *et al.*, 2011). A number of studies attempted to estimate alpha and beta diversity by relating the spectral variation of a site to the ecosystems' heterogeneity at different spatial scales and in different habitat types (Möckel *et al.*, 2016; Rocchini *et al.*, 2010). The reasoning behind this approach is that environmental heterogeneity and high biological diversity are interconnected, because heterogeneous areas are likely to harbour more species due to a higher number of available ecological niches. By regressing field data on the distribution of plant strategies, Schmidtlein (2012) developed models and applied them to airborne hyperspectral imagery on a per pixel basis. The resulting local maps demonstrated the potential to detect community strategy type composition and showed ways to interpret them in terms of plant species composition and environmental constraints. As the aforementioned three strategies are related to the levels of productivity and disturbance at a given site, their change in space and time may serve as a measure of key processes such as succession, eutrophication and other changes in habitat conditions and may provide direct insights into the spatial ecology of a grassland area (Schmidtlein *et al.*, 2012). Ellenberg indicator values for water supply, soil pH and soil fertility from montane rangelands were regressed to reflectance values extracted from airborne hyperspectral imagery (Schmidtlein, 2005). When applying the regression to the imagery, largely accurate maps could be produced giving the spatial distribution of soil attributes as indicated by the Ellenberg values (R^2=0.58-0.68 in cross-validation), which makes them an appealing tool for vegetation monitoring.

Synthetic aperture radar (SAR) and light detection and ranging (LIDAR)

Over recent years, there has been increasing interest in synthetic aperture radar (SAR) data for aboveground biomass analyses, particularly in the areas of frequent cloud conditions where obtaining high quality optical data is difficult. The capability of radar systems to collect data in all weather and light conditions overcomes this issue. Furthermore, the SAR sensor can penetrate vegetation to different degrees and provides information on the amount and three-dimensional (3-D) distribution of structures within the vegetation. The basic operating principle of the radar system includes the transmittance of microwave energy (wavelengths within the approximate range of 1 mm to 1 m) from an antenna in very short bursts or pulses. By electronically measuring the return time of signal echoes, the range, or distance, between the transmitter and reflecting objects may be determined. Vegetation with high moisture content returns more energy than dry vegetation, and more energy returns from crops having their rows aligned in the azimuth direction than from those aligned in the range direction of radar sensing. Radar based sensors are active and function independently of solar radiation variations, unlike optical sensors which depend on solar radiation. However, radar use has limited applications in regional or small-scale studies due to the small swath width, high costs of airborne acquisitions, lower sampling density of the large footprint waveform, and a limited extent of coverage. LiDAR, like radar, is an active RS technique. This technology involves the use of pulses of laser light directed towards objects and measuring the time of pulse return, which is processed to calculate the distances between the sensor and the various objects. LIDAR systems have the capability to capture reflectance data from the returning pulses, in addition to the three-dimensional coordinates of the returns. Commercial LIDAR systems frequently utilise a rapidly pulsing laser (up to 70,000 pulses sec^{-1}) with a 1064 mm near-infrared wavelengths pulse. Such systems also allow the recording of the intensity of LIDAR echoes, which varies with the wavelength of the source energy and the composition of the material returning the incoming signal.

With the aim to monitor grassland by the use of multi-temporal optical and radar satellite images, Dusseux *et al.* (2014) showed that SAR images allowed a better discrimination than optical images between grasslands and crops in agricultural areas where cloud cover is very high most of the time, which restricts the use of visible and near-infrared satellite data. The results show that the classification accuracy of SAR variables was higher than those using optical data (R^2 of 0.98 compared to 0.81). McNairn *et al.* (2008) demonstrated that multi-temporal SAR imagery can successfully classify crops for a variety of cropping systems present across Canada. Overall accuracies of at least 85% were achieved, and most major crops were also classified to this level of accuracy. Several studies have established a strong correlation between LiDAR metrics and aboveground biomass. However, most of the studies were conducted in forests or savannas and grasslands with substantial wood encroachment (McGlinchy *et al.*, 2014) which are characterized by an uneven distribution of vegetation biomass in 3-D space with biomass allocated to above and below ground components. Furthermore, the structure of savanna vegetation is variable with the occurrence of an herbaceous layer with variable tree cover and open spaces. These structures differentiate them strongly from typical temperate grasslands, which are less heterogeneous and rarely show tree or shrub encroachment. Although LiDAR data have some advantages over optical data, there are a few issues that restrict its use for field applications. For example, LiDAR data analyses are not simple and require more image processing knowledge and skill and specific software (Kumar *et al.*, 2015). The LiDAR data acquisition process is expensive and covers smaller area, hence, study areas are still limited to specific areas and have not been applied extensively to larger areas for biomass estimation. Despite the popularity of radar and LiDAR data in forest biomass analyses, no studies exist up to now which have utilized such data in the estimation of temperate grassland biomass.

Ultrasound

Ultrasonic sensors determine the distance to/from an object by recording the time difference between the transmission of an ultrasonic signal (burst) and the reception of the signal's echo reflected by the object. Commercial sensors often utilize a one-headed system with one sonic transducer (frequency approx. 180 kHz) that acts both as transmitter and as receiver (Pepperl and Fuchs, 2010). Ultrasonic sensors have been used since the late 1980s in tree canopy height and volume measurements (Lee *et al.*, 2010). These sensors are widespread in process applications (Hauptmann *et al.*, 1998) and can provide high efficiency at low cost (Park *et al.*, 2010). Although the accuracy of modern ultrasonic sensors has improved, difficulties in interpreting the data often occurs due to variance in measurement conditions and transducer behavior (Henning *et al.*, 2000). Across a biomass range of 0.35-2 t ha^{-1} in areas continuously grazed by sheep, measurements with an ultrasonic sensor underestimated sward height using top canopy heights as reference. In spite of this, biomass estimations were promising having R^2-values between 0.66 and 0.81 (Hutchings *et al.*, 1990). Sonic reflections for ryegrass-dominated swards were partly weak due to erect leaf orientation. The complex interaction between sward structure and reflection from the ultrasonic sensor is significantly affected by size, angle, and surfaces of leaves. Sensor-specific effects also play a role in the interaction (Hutchings, 1991, 1992). Having installed the sensor on a tractor, Scotford and Miller (2004) conducted ultrasonic sensor on-the-go studies in different winter wheat varieties with erect leaf canopies. Deviations between 4.6 and 7.2 cm from the reference crop height values were obtained. Reusch (2009) used a specific configuration of an ultrasonic sensor with an adapted control unit to estimate biomass in winter wheat. With this system, it was possible to retrieve multiple echoes from different leave layers and from the ground and thus was independent of the sensor's mount height. Forage mass-height relationships were evaluated by carrying out static ultrasonic measurements on binary legume-grass mixtures of white clover (*Trifolium repens* L.), red clover (*Trifolium pratense* L.), and lucerne (*Medicago sativa* L.) with perennial rye grass (*Lolium perenne* L.) across a wide range of sward heights and forage mass (Fricke *et al.*, 2011). A common calibration model for aboveground biomass including all sward types based on ultrasonic sward height explained 74.8% of the variance with a standard error (SE) of 1.05 t ha^{-1}. Contrary, in heterogeneous pastures, which showed a wide variation in species composition, phenological stage and sward architecture, ultrasonic recordings showed limited accuracy when correlated with grassland biomass (Safari *et al.*, 2016b).

Sensor combinations

Most studies involving biomass estimation from RS data have used a single sensor or single date image, which may not be sufficient for complex applications such as biomass estimation in certain areas or in grasslands with high botanical diversity and structural diversity. Since RS data are available from a range of sensors, each with its own characteristics, a combination of sensors may be beneficial to provide a better information on the observed stand. Some information exists on the integration of multiple sensors for the estimation of aboveground biomass in forests. For example, the combination of spectral (i.e. data from Landsat ™) and spatial (i.e. radar data) information contents improved model performance for forest area (Haack *et al.*, 2002). Contrary, for grassland very little research exists on the benefit of sensor integration. Recent studies have shown that the fusion of optical and ultrasonic data resulted in an improved performance for biomass and quality estimation of highly heterogeneous pastures (Safari *et al.*, 2016a, b). Combining ultrasonic sward height data with narrow band normalized spectral vegetation index (NDSI) or WorldView2 satellite broad bands (WV2) reduced the standard error of cross validation (CV) for grassland biomass by up to 39% compared to the use of exclusive sensors. These combinations can be on a par or even better than the use of the full hyperspectral information. Narrow-band NDSI constructed with bands in these spectral regions may be preferred for research purposes where highest accuracy is essential, whereas WV2 provides interesting perspectives for practical implementation, as these bands are already implemented on satellite platforms. The same sensor combination increased the prediction accuracy for dry matter yield of pure reed canary grass swards, white clover/grass and high diversity mixtures (R^2_{cv}=0.79) compared to the exclusive use of ultrasonic sensor (R^2_{cv}=0.73) and NDSI (R^2_{cv}=0.38) (Reddersen *et al.*, 2014). However, the inclusion of leaf area index in a triple sensor approach further improved the accuracy (R^2_{cv}=0.81), which indicates that even dual sensor systems may not fully exploit the potentially available stand information. Further, the presence of a high proportion of senesced material in pastures influences the performance of the sensor systems and may limit the applicability of such concepts in situations with advanced canopy age e.g. with low stocking rates. Thus, more advanced sensor systems and methods are required to overcome the existing limitations.

Conclusions

Grassland systems frequently exhibit a small-scale botanical and structural heterogeneity with pronounced spatio-temporal dynamics. Such features pose particular challenges for any sensor applications. Regarding these problems, the considerable costs and frequently limited spatial resolutions of many available RS systems, it is comprehensible that up to date hardly any commercial application of RS exists for practical grassland farming in Europe. However, considering the developments in sensor performance, data processing and analysis and modelling over the recent decades, there is scope for significant advances in RS for grassland research and practice. It is mainly improvements in spatial resolution, which will greatly promote applicability of RS. Further increases in spectral resolution can be expected, which will increase the accuracy of spectral measurements. Development of systems with complementary sensors are seen as a most promising research area, which will help to overcome the limitations of single sensors and to provide better information about grassland composition, yield and quality.

From an agronomic point of view, the central outcome of RS and data analysis are thematic maps from farm fields, which represent the relevant grassland features and constitute the basis for various farm management measures, i.e. on the:

1. Strategic level, where long-term decisions are taken based on aggregated data over time for the farm and on future scenarios created from down-scaled climate scenarios (e.g. farm infrastructure planning).
2. Tactical level, where medium-term decisions are made (e.g. evaluation of clover dry matter contribution in pastures and choice of crop species for oversowing).
3. Operational level, where farmers take day-to-day decisions based on spatially explicit real-time data on yield and quality of pastures (e.g. planning of ration, pasture rotation and fertilizer application).

Eventually, the overarching goal will be to provide cheap, appropriate and timely information to farmers to support farmer's decision-making.

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Effect of wilting and silage additives on silage quality of lucerne, red clover and legume-grass mixtures

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Abstract

Lucerne (*Medicago sativa*), red clover (*Trifolium pratense*) and the mixtures with grass are regarded difficult to ferment based on low contents of water-soluble carbohydrates (WSC) and nitrate as well as high buffering capacity (BC). The objective of this laboratory ensiling trial was to investigate the effects of 24, 48 and 72 hours wilting on dry matter (DM)-, WSC-content, BC as well as content of yeasts and lactic acid bacteria (LAB). Furthermore, the effects of chemical and biological additives on silage quality were investigated regarding the concentration of lactic acid, acetic acid and butyric acid, ethanol and ethyl esters depending on the level of wilting in ensiling material. In total 144 laboratory silages were analysed. With extended wilting period, the contents of WSC and BC decreased, whereas the contents of yeasts and LAB increased. A lower range of DM is associated with a higher risk of butyric acid formation, which requires the addition of chemical additives against clostridia activity. The results demonstrate the occurrence of volatile organic compounds (VOC), especially ethanol and ethyl esters, in legume silages. The DM content and silage additives affect the concentrations of ethanol, acids and esters. Elevated levels of ethanol and esters occur in silages with low DM. For the reduction of ethanol and ester formation, chemical additives against yeasts and bacteria are necessary.

Keywords: legumes, silage quality, volatile organic compounds, esters

Introduction

Legumes are crops difficult to ensile due to low contents of water-soluble carbohydrates (WSC), buffering capacity (BC) and nitrate. However, they play an important role in supplying nitrogen in rotational food production systems and may have a high nutritive value, especially high contents of crude protein. The objective of this study was to examine the silage management factors such as wilting and silage additives on legume and legume/grass mixture silage quality in combination with the incidence of VOC. Weiß and Auerbach (2013) postulated that esters can be frequently found in maize, whole crop wheat and sorghum silages and also in grass silages and that ester concentrations are strongly correlated with the ethanol concentration and silage pH.

Materials and methods

Lucerne, red clover and the mixtures with grass were harvested on 1 June 2015 near Oldenburg, Germany, chopped to a theoretical particle length of 4 cm and wilted for 24, 48 and 72 hours. Fresh and wilted ensiling material (EM), with and without any additives, were filled into 1.5 L glass jars and stored for 90 days under anaerobic conditions at 20 °C. All treatments were done in triplicates. Each crop received the ensiling treatments of no additive (Control), different commercial products of lactic acid bacteria (LAB 1, LAB 2) with inoculation rates of 10^5 cfu g^{-1} FM, and commercial chemical products with a mixture of nitrite, hexamine and/or benzoate (Salt). Herbage (EM) with DM contents of <400 g kg^{-1} was treated with products of aim-of-acting (WR) 1a, 1b or 1c according to DLG (2011), EM with DM >400 g kg^{-1} with products of WR 2. The contents of DM, LAB, yeasts, WSC, BC and nitrate were analysed according to standard methods (LUFA). Silage analyses were conducted in aqueous extracts of frozen silages as described by Weiss and Auerbach (2013). Lactic acid (LA) was analysed by HPLC.

Fermentation acids, alcohols and esters were determined by GC. Statistical analyses were performed with SAS 9.3. For treatment comparisons, the analysis of variance was used.

Results and discussion

The legumes used in the trial (Table 1) were estimated as difficult (fermentation coefficient FC <35) or moderate fermentable (FC 35-45) according to DLG (2011). Three EM with FC >45, all wilted for 72 h, had DM contents over 400 g kg^{-1} and low levels of WSC. Although all EM were nitrate-free and thus with a higher risk for poor fermentation, butyric acid (BA) was only analysed in lucerne after 48 h of wilting and DM of 266 g kg^{-1} (data not shown). BA could be suppressed only with silage salt (WR 1a, DLG (2011)). Polyphenol oxidases (PPO), which occur in red clover (Jones *et al.*, 1995), are able to inhibited proteolysis. On the other hand high levels of DM and therefore higher a_w-values in combination with higher acidification rate can reduce clostridia activity (Kaiser *et al.,* 2009). The epiphytic content in our study was high (Table 1) and increased with wilting period. Lactic acid contents ranged between 49.8 and 104.5, acetic acid between 11.8 and 26.8 g kg^{-1} DM (data not shown). Laboratory ensiling trials are characterised by clean experimental conditions that means free of clostridia spores. However, as shown in Table 1, yeast counts were high and increased during wilting period. In accordance to the fact that under anaerobic conditions yeasts are responsible for ethanol formation, the ethanol content in silages without additives was between 4.8 and 10.9 g kg^{-1} DM (Table 2) with a strong negative correlation to DM content (R^2=0.81) and positive correlation to ester content (R^2= 0.65). The total esters ranged between 124 and 197 mg kg^{-1} DM in untreated silages and consisted of only ethyl lactate. These ester contents are comparable with contents in grass silage (Weiß and Auerbach, 2013) considering the pH level between 4.0 and 6.3. As shown in Table 2 the contents of ethanol were mainly not affected by silage additives with LAB, the same applies for the contents of esters. The additive salts containing benzoate, nitrite and hexamine strongly reduced the ethanol and ester contents. According to Woolford (1975) these substances are able to inhibited yeasts and possibly heterofermentative LAB which also produce ethanol.

Conclusions

Ensiling within the lower DM range of legumes is associated with a higher risk of BA formation in lucerne. Elevated levels of ethanol, particularly in the lower range of DM, and esters occur in legume silages. For the reduction of ethanol and ester formation, chemical additives against yeasts and bacteria are necessary.

Table 1. Effect of wilting period (WP) on DM, parameters of ensilability and microbial composition in legumes.[1]

Legume	WP	DM g kg^{-1} FM	WSC g kg^{-1} DM	WSC/BC	FC FC= DM(%) + 8WSC/BC	LAB log cfu g^{-1} FM	Yeasts log cfu g^{-1} FM
Lucerne (Lu)	24 h	232	39	0.6	28	7.2	6.8
48 h		266	0	0	27	8.2	8.4
72 h		471	2	0	47	8.1	8.4
Red clover (Rc)	24 h	179	89	1.7	32	6.9	7.3
48 h		209	26	0.4	24	7.7	8.1
72 h		333	11	0.2	35	7.7	8.1
Mixture of Lu/ Grass	24 h	245	75	1.5	36	7.7	7.5
48 h		316	33	0.6	36	8.6	7.6
72 h		416	33	0.6	46	8.4	7.8
Mixture of RC/ Grass	24 h	211	107	2.3	39	7.1	7.3
48 h		284	51	0.9	35	7.8	8.3
72 h		440	30	0.6	49	7.8	8.1

Table 2. Effects of silage additives on contents of ethanol and esters in silages of legumes.[1]

Legume/ WP		Control	LAB1	LAB2	Salt	SEM	P[2]
		Ethanol					
Lucerne (Lu)	24 h	10.8^{a}	8.8^{ab}	8.2^{b}	3.2^{c}	0.5	<0.001
	48 h	9.0^{a}	8.6^{a}	8.4^{a}	1.8^{b}	0.3	<0.001
	72 h[3]	4.8^{a}	4.7^{a}	4.8^{a}	1.3^{b}	0.03…0.2	<0.001
Red clover (Rc)	24 h	10.7^{a}	9.9^{a}	9.8^{a}	1.6^{b}	0.5	<0.001
	48 h	11.1^{a}	10.9^{a}	11.0^{a}	2.0^{b}	0.2	<0.001
	72 h[3]	7.5^{a}	7.7^{a}	7.3^{ab}	1.8^{b}	0.1…0.9	0.047
Mixture of Lu/Grass	24 h	8.9^{a}	9.2^{a}	8.4^{a}	2.0^{b}	0.3	<0.001
	48 h	7.4^{a}	7.6^{a}	7.3^{a}	2.6^{b}	0.9	0.002
	72 h[3]	5.1^{b}	6.2^{a}	5.5^{b}	1.5^{c}	0.2	<0.001
Mixture of RC/Grass	24 h	8.9^{a}	8.7^{a}	8.6^{a}	1.6^{b}	0.3	<0.001
	48 h	10.9^{a}	10.9^{a}	10.8^{a}	1.3^{b}	0.3	<0.001
	72 h[3]	6.3^{a}	6.8^{a}	5.4^{a}	1.0^{b}	0.3	<0.001
		Ester total (ethyl acetate + ethyl lactate)					
Lucerne (Lu)	24 h	142^{a}	129^{b}	126^{b}	0^{c}	2.7	<0.001
	48 h	130^{ab}	129^{ab}	n.a.[4)]	0^{b}	6.5…64.4	0.003
	72 h[3]	155^{a}	126^{a}	100^{b}	67^{b}	1.5…16.9	0.017
Red clover (Rc)	24 h	182^{a}	190^{a}	189^{a}	0^{b}	5.8	<0.001
	48 h	166^{a}	171^{a}	168^{a}	0^{b}	2.8	<0.001
	72 h	148^{a}	177^{a}	143^{a}	0^{b}	27.6	0.008
Mixture of Lu/Grass	24 h	189^{a}	137^{b}	119^{b}	83^{c}	7.7	<0.001
	48 h	124^{a}	126^{a}	116^{b}	0^{c}	1.3	<0.001
	72 h[3]	130^{ab}	222^{a}	184^{a}	0^{b}	39.5	0.019
Mixture of RC/Grass	24 h	191^{ab}	202^{a}	185^{b}	0^{c}	2.5	<0.001
	48 h	197^{a}	206^{a}	205^{a}	0^{b}	4.0	<0.001
	72 h[3]	171^{a}	156^{a}	175^{a}	0^{b}	5.4	<0.001

[1] n=3; means within rows are significantly different if they have no letters in common ($P<0.05$, Tukey`s test).

[2] P-values of global F-test.

[3] Silage additives according DLG aim-of-acting 2, otherwise DLG 1b (LAB1) or 1b, c (LAB 2).

[4] Not analysed.

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Dry matter losses and hygienic quality of grass silage inoculated with different lactic acid bacteria strains

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Abstract

The current study focuses on the effects of five inoculants containing various combinations of novel lactic acid bacteria strains on fermentation end products, microbial composition and aerobic stability of grass silage. All inoculants were applied at an application rate of 150,000 cfu g^{-1} forage. After ensiling period of 90 d, silage was sampled and analysed for dry matter (DM), lactic acid and volatile fatty acids (VFA), ethanol, ammonia-N, number of clostridia spores, yeasts and moulds. The DM and DM losses were corrected for volatiles. Aerobic stability of the silages was measured as number of days reaching a temperature of 3 °C above ambient temperature. Data were analysed as a randomized complete block design by using Proc GLM of SAS with treatment as a fixed factor. Five replications per treatment were used. Selected lactic acid bacteria inoculants improved fermentation characteristics, aerobic stability and hygienic quality of grass silage compared to spontaneous fermented silage. Single heterofermentative strain of *Lactobacillus buchneri* or blends of hetero-and homofermentative lactic acid bacteria were more effective in improving aerobic stability.

Keywords: lactic acid bacteria, silage, aerobic stability, mould, yeast

Introduction

Silage quality and nutrient use efficiency is influenced by a number of factors: such as crops, ensiling technologies, used machinery and additives for manipulating fermentation processes (Davies *et al.*, 2005). Microbial additives containing lactic acid bacteria (LAB) are commonly used for silage preservation to achieve a rapid pH drop through organic acid production, and some strains have demonstrated their efficacy to improve aerobic stability, by inhibiting spoilage moulds and yeasts (Muck, 2012; Tabacco *et al.,* 2011).Silage research has traditionally focused on the production and ensiling of grass and legume silages with references to improving fermentation, aerobic stability and hygienic quality, and reduction of DM losses. The current study focuses on the effects of inoculants containing various combinations of novel bacterial strains on fermentation end products, aerobic stability and DM losses of grass silage.

Materials and methods

A grass mixture (70% perennial ryegrass (*Lolium perenne* L.) and 30% timothy *(Phleum pratense* L.), wilted to a DM content of 26.5%, was precision chopped and ensiled in 3 litre laboratory silos. The forage was T1 – untreated or treated with T2 – *Lactobacillus buchneri*; T3 – *Lactobacillus plantarum*, *Enterococcus faecium* and *Lactobacillus buchneri*; T4 – *Lactobacillus plantarum*, *Enterococcus faecium* and *Lactococcus lactis*; T5 – *Enterococcus faecium*, *Lactococcus lactis* and *Lactobacillus plantarum*; T6 – *Lactobacillus plantarum* and *Lactobacillus plantarum*. All inoculants were applied at a rate of 150,000 cfu g^{-1} forage. Silages were analyzed on day 90 of storage at 20 °C for DM, lactic acid and VFA, ethanol, ammonia-N, number of clostridia spores, yeasts and moulds, and an aerobic stability lasting for 13 days. Aerobic stability was defined as the number of hours the silage remained stable before rising more than 3 °C above the ambient temperature. Silage composition data and aerobic stability data were analyzed using Proc GLM of SAS, version 8.02, 2000 and LSD tests were used to indicate significant differences between untreated and additive treatment. Significance was declared at $P<0.05$.

Results and discussion

The quality of inoculated silages was significantly increased compared to that of the untreated, control silage (Table 1). Use of this products resulted in higher DM concentration lower DM loss compared with the untreated silage. The T5 silage had the lowest (P<0.05) DM loss. All five products resulted in significantly (P<0.05) lower pH reduction after 3 d and 90 d of fermentation compared with the untreated silage. Filya *et al.* (2007) concluded that the main effects of silage inoculants were increased production of lactic acid connected with significant reduction of the pH value and minimised DM losses. When compared with untreated silage, increased (P<0.05) acetic acid concentration and decreased lactate:acetate ratio were observed only in the T2 silage. The treatments T3-T6 produced higher (P<0.05) lactic acid concentration and higher lactate:acetate ratio compared with the untreated and T2 silage. The acetic acid concentration was highest in the T2 silage. The inoculation resulted in lower proteolysis compared to control silages as the inoculated silage had significantly lower concentration of ammonia-N. The inoculated silages also had lower (P<0.05) concentrations of alcohols and butyric acid compared to control silage. The LAB blends used in our experiment significantly suppressed yeast and mold growth and was reflected in a lower concentration of alcohols compared with control silage, generally correlated to yeast activity in silage.

The highest aerobic stability and lowest yeast and mould counts were found in T2 and T3 silages (Table 1). The improved stability was related to the lowest pH value after aerobic exposure for 7 days. The aerobic stability of T4-T6 silages was improved by 2.7 d (66 h), the aerobic stability of T3 silages was improved by 5.7 d (138 h) compared to control silages. The aerobic stability of T2 silages was improved drastically (Table 1; Figure 1). Danner *et al.* (2003) provide evidence for the existence of certain LAB strains with the power to inhibit yeasts and mold growth and to improve aerobic stability.

Table 1. Results of bacteria-strain treatment of an ensiled ryegrass:timothy sward. Dry matter, calculated dry matter losses and fermentation parameters are corrected for volatiles.[1,2]

	T1	T2	T3	T4	T5	T6	$LSD_{0.05}$
Corr. DM, g kg^{-1} FM	250^{c}	$252^{b,c}$	$252^{b,c}$	$254^{b,a}$	256^{a}	$254^{b,a}$	2.56
DM loss, g kg^{-1} DM	63^{a}	$58^{b,a}$	$52^{b,c}$	$45^{d,e}$	40^{e}	$46^{d,e}$	6.71
pH after 3 days	5.48^{a}	4.90^{b}	4.38^{e}	4.70^{c}	$4.45^{d,e}$	$4.59^{d,c}$	0.142
pH after 90 days	4.55^{a}	4.15^{b}	3.89^{d}	3.98^{c}	3.96^{c}	3.91^{d}	0.037
pH after aerobic stability test	8.81^{a}	4.43^{e}	4.76^{d}	$8.64^{c,b}$	8.70^{b}	$8.62^{c,b}$	0.104
Ammonia-N, g kg^{-1} total N	60^{a}	51^{b}	49^{b}	43^{c}	41^{c}	43^{c}	4.67
Lactic acid, g kg^{-1} DM	31^{d}	34^{d}	53^{c}	58^{c}	58^{c}	65^{b}	5.99
Acetic acid, g kg^{-1} DM	$28^{c,b}$	46^{a}	$26^{c,b}$	$25^{c,d}$	28^{b}	24^{d}	3.53
Butyric acid, g kg^{-1} DM	2.5^{a}	$1.9^{a,b}$	1.6^{b}	0.6^{c}	0.4^{c}	0.7^{c}	0.76
Propionic acid g kg^{-1} DM	$0.16^{b,a}$	0.28^{a}	$0.22^{b,a}$	0.12^{b}	0.14^{b}	0.10^{b}	0.127
Alcohols, g kg^{-1} DM	7.0^{a}	5.9^{b}	$5.5^{c,b}$	4.8^{c}	$5.0^{c,b}$	4.9^{c}	0.94
Clostridia, log cfu g^{-1}FM	0.99^{a}	0.99^{a}	0.99^{a}	0.99^{a}	0.99^{a}	0.99^{a}	0.004
Yeast, log cfu g^{-1}FM	3.2^{a}	1.3^{b}	1.4^{b}	1.6^{b}	1.5^{b}	1.4^{b}	0.61
Mould, log cfu g^{-1}FM	3.0^{a}	1.3^{b}	1.4^{b}	1.6^{b}	1.4^{b}	1.7^{b}	0.39
Aerobic stability, h	60^{b}	150^{a}	138^{a}	144^{a}	78^{c}	96^{c}	3.06

[1] Means with different superscript letters within a row indicate significant differences of P<0.05. $t_{0.05}$ = 2.0639; Error df = 24.

[2] T1-T6 = See text in the Materials and methods for explanation of treatments T1-T6.

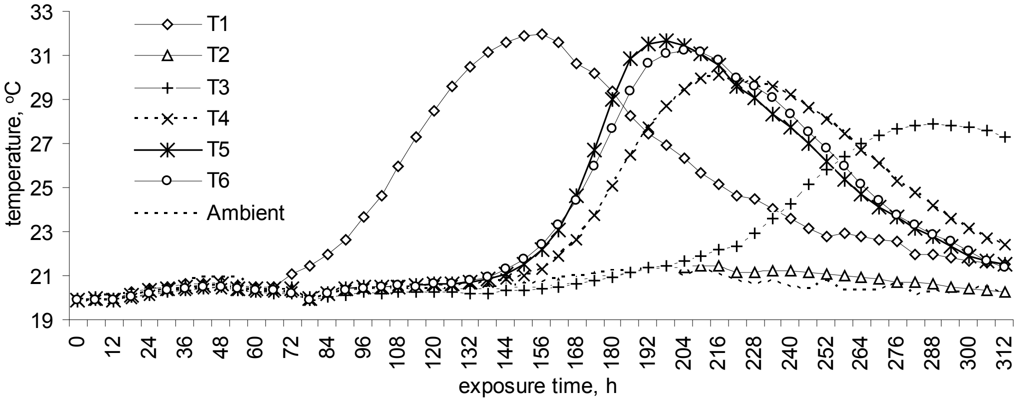

Figure 1. Temperature changes in the perennial ryegrass/timothy silage during aerobic exposure. T1-T6 = see text in the Materials and methods for explanation of treatments T1-T6.

Conclusions

In this study selected lactic acid bacteria inoculants improved fermentation characteristics, aerobic stability and hygienic quality of grass silage compared to spontaneous fermented silage. A single heterofermentative strain of *Lactobacillus buchneri* or blends of hetero- and homofermentative lactic acid bacteria were more effective in improving aerobic stability.

Acknowledgements

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Effect of mower/conditioner type during legume or grass harvesting on silage quality

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Abstract

Machinery for harvesting forage is typically designed for use with grass crops and often incorporates conditioners to promote moisture loss during wilting. Forage legumes have fragile leaves that can shatter during conditioning treatment and since leaves contain higher concentrations of protein than stem, reducing leaf shatter should increase the forage protein yield at harvest. This study examined the effects of different mower treatments on silage prepared from ryegrass (*Lolium perenne*), red clover (*Trifolium pratense*), white clover (*Trifolium repens*) or lucerne (*Medicago sativa*). Three mower treatments were compared: no conditioner (NoC), with a steel tine conditioner (STC) or a with rubber roller conditioner (RRC). The RRC improved wilting rate producing higher dry matter silages than when no conditioner was used, but lower DM silages than when the STC was used. Silage produced using a STC had a lower nitrogen content than silage produced using NoC or a RRC suggesting that a RRC was preferable to the STC when harvesting legumes so as to limit loss of nitrogen rich leaf material.

Keywords: legumes, nitrogen, wilting, conditioning, silage

Introduction

Forage crops should be wilted rapidly to ensure that nutrient loss is kept to a minimum during ensiling and forage conditioners are often used during forage harvesting to achieve rapid wilting. However, the farm machinery for harvesting forage in the UK has typically been designed for use with grass rather than legume forages. Forage legumes have fragile leaves that can shatter during conditioning treatment and since these leaves contain higher protein concentrations than the stem (Grewal and Williams, 2002), reducing leaf shatter should increase protein yield. Research was needed to determine the potential losses caused by different mower conditioners when harvesting legume crops. The aim of this experiment was to compare four forages: red clover; white clover; lucerne or perennial ryegrass, harvested and ensiled to evaluate the effect of different conditioners on nutrient quality of the forage and silage.

Materials and methods

Triplicate plots (13.5×11 m) of white clover (cv. Aran; WC), red clover (cv. AberClaret; RC), lucerne (cv. Timbale; LUC) and perennial ryegrass (cv. AberMagic; PRG) were established in a randomised complete block (RCB) design in June 2013. In the 2nd cut of 2014, three mower treatments were investigated in a split plot design, each mower cutting a 9×3 m strip within each forage plot. Mower treatments were: (1) no conditioner; (2) standard steel-tine conditioner; and (3) rubber roller conditioner. The mowers used were Novacat models 301, 301 ED and 301 RC respectively (supplied by Alois Pottinger UK Ltd., Corby, UK). All mowers were front mounted 3 m-wide machines set to an 8 cm cutting height and to form a 1.6 m wide swath. After a 24 h wilt and without further movement, the forage was baled using a fixed chamber round baler (Model 568, John Deere Ltd, Langar, Notts., UK). The baled forage was removed from the baler and a 1.5 kg sample was taken using a 50 mm Ø corer. A subsample of the core was ensiled in a 1.5 l glass laboratory silo after treatment with an inoculant (Ecosyl 100, Volac Ltd, Royston, Herts., UK; 1×10^6 cfu g^{-1}). Samples of standing crop, baled forage as ensiled and the silages (n=36) were taken for dry matter (DM) determination at 100 °C and further samples were freeze dried prior to determination of ash, water soluble carbohydrate (WSC), nitrogen (N), neutral-detergent fibre (NDF) and digestible organic matter in the DM (DOMD) content. Ammonia-N, pH and lactate content was determined on fresh frozen silage samples. Ensiled forage/silage and standing forage data were analysed by ANOVA as split plot and RCB designs respectively.

Results and discussion

In the standing crops, DM content was highest in PRG and lowest in WC with RC and LUC intermediate (Table 1). N content was lower in PRG than in legumes and higher in WC than RC. WSC was highest in PRG and lowest in LUC. At ensiling, PRG DM content was higher than in RC or WC with LUC intermediate. N content of PRG was lower than in legumes and RC was lower than WC and LUC. Forage harvested using the rubber roller conditioner (RRC) had higher DM than that harvested with NoC but lower DM than with the steel tine conditioner (STC).

PRG silage DM content was higher than both RC and WC and LUC was higher than WC. N concentration of PRG silage was lower than in the legumes and RC was lower than WC and LUC. As seen in the forage pre-ensiling, silage harvested using the RRC had a higher DM than forage harvested

Table 1. Composition of standing crop, forage as ensiled and silage composition after 143 days (g kg^{-1} DM unless otherwise stated).[1,2,3]

		Forage (F)				Mower (M)			s.e.m.		Prob	
		PRG	RC	WC	LUC	NoC	STC	RRC	F	M	F	M
Standing crop	Oven DM g kg^{-1}	246^{b}	200^{ab}	180^{a}	214^{ab}	–	–	–	12.1	–	0.040	–
	Ash	53^{a}	62^{b}	73^{c}	66^{bc}	–	–	–	2.0	–	0.002	–
	Nitrogen	16.2^{a}	24.2^{b}	32.2^{c}	28.5^{bc}	–	–	–	1.44	–	0.001	–
	WSC	292^{c}	160^{b}	158^{b}	90^{a}	–	–	–	12.6	–	<0.001	–
	NDF	453^{c}	365^{b}	311^{a}	449^{c}	–	–	–	9.5	–	<0.001	–
Forage as ensiled	Oven DM g kg^{-1}	399^{b}	323^{a}	312^{a}	379^{ab}	327^{A}	378^{C}	355^{B}	15.5	7.9	0.018	0.001
	Ash	62^{a}	75^{b}	86^{c}	74^{b}	74	74	75	0.5	0.8	<0.001	0.758
	Nitrogen	19.0^{a}	27.3^{b}	32.4^{c}	32.8^{c}	28.2	27.8	27.6	1.11	0.66	<0.001	0.770
	WSC	242^{c}	142^{b}	130^{b}	78^{a}	128	128	124	$0.00044^{\$}$	$0.00013^{\$}$	<0.001	0.358
	NDF	515^{d}	409^{b}	351^{a}	462^{c}	436	433	435	6.9	5.7	<0.001	0.915
Silage	Oven DM§ g kg^{-1}	351^{c}	290^{ab}	276^{a}	349^{bc}	291^{A}	340^{C}	318^{B}	17.0	6.8	0.041	<0.001
	Ash	67^{a}	77^{b}	90^{c}	75^{b}	77	77	79	1.4	0.9	<0.001	0.194
	Nitrogen	19.6^{a}	29.0^{b}	34.8^{c}	33.9^{c}	29.8^{B}	28.8^{A}	29.5^{B}	0.64	0.23	<0.001	0.022
	NDF	503^{d}	420^{b}	345^{a}	464^{c}	428	438	432	10.2	4.5	<0.001	0.312
	DOMD	718^{c}	641^{b}	696^{c}	597^{a}	667	661	660	9.7	5.5	<0.001	0.640
	WSC	200^{c}	56^{b}	54^{b}	19^{a}	57^{A}	65^{B}	54^{A}	0.058^{5}	0.016^{5}	<0.001	0.009
	pH	3.77^{a}	3.79^{a}	3.77^{a}	4.07^{b}	3.83	3.87	3.86	0.052	0.025	0.018	0.558
	Lactate	67^{a}	96^{b}	99^{b}	72^{a}	88^{B}	78^{A}	84^{B}	4.3	1.9	0.004	0.009
Ammonia-N	NoC	84	57	77	$100^{a'}$			F	0.0332^{4}		0.040	
(g kg^{-1} N)	STC	78	57	78	$78^{b'}$			M	0.0077^{4}		0.038	
	RRC	74	59	86	$83^{b'}$			F.M	0.0154^{4} , $0.0355^{4,5}$		0.011	

[1] Within rows and either forage or mower treatments differing superscripts indicate means differ (*P*<0.05) based on Student-Newman-Keul's method except § Fisher's protected LSD. s.e.m. applies to $^{\$}$reciprocal of means or $^{\#}$means on $\log_{10}$ scale.

[2] a,b; means differ (*P*<0.05) based on Bonferroni method.

[3] DM = dry matter; NDF = neutral detergent fibre; DOMD = digestible organic matter in the DM; WSC = water soluble carbohydrates; NoC = no conditioner; STC = steel tine conditioner; RRC = rubber roller conditioner; PRG = perennial rye grass; RC = red clover; WC = white clover; LUC = lucerne.

[4] s.e.m. applies to Box-Cox transformed means y = (-0.2x – 1)1/-0.2.

[5] Within-F interaction s.e.m. on 7 df.

with NoC and a lower DM than the STC. Although forage pre-ensiling showed no difference in N amongst mower treatments, silage harvested using a STC had lower N than both other mower treatments, indicating greater loss of protein rich leaf during harvest (Fychan *et al.*, 2015). Differences in fermentation parameters were observed between forages, with a higher pH observed in LUC compared with the other forages. Lactate levels were lower in PRG and LUC and consistent with their higher DM contents.

The higher DM content observed in the STC treatments resulted in higher residual WSC and lower lactate concentrations when compared to the other mower treatments. Mower type had no effect on ammonia-N levels in PRG, RC and WC silages but levels were lower in conditioned rather than unconditioned LUC which may reflect its lower DM content and limited WSC and consequent higher pH.

Conclusions

Using a rubber roller conditioner increased forage wilting rate compared to the no conditioner treatment but it did not increase wilting rate as effectively as the steel tine conditioner. Overall, the N concentration of the rubber roller and no conditioner silages were similar and higher than those from a steel tine conditioner.

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Hay sampling methods affect the results of forage analyses

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Abstract

Accurate and representative sampling of stocked hay is crucial to obtain reliable results of laboratory analyses of forage quality. Although the use of coring devices is advisable, hand-grabbing is very common among farmers. In order to elucidate the effect of these two sampling methods on the results of the forage analyses, a field experiment was conducted in South Tyrol (Italy). Grab samples were obtained at 10 to 15 spots of the upper surface of piles of loose hay stored in barns. Core samples were obtained at three randomly selected spots using a modular, mechanical coring device with a 2.25 cm inner diameter and a sampling depth up to 3.5 m. Samples were taken with both sampling techniques at 48 farms. Hay from the first cut as well as hay from the second cut was sampled at each farm. Hand-grabbing negatively affected several parameters of forage quality, resulting in a decrease of ash and crude protein content and in an increase of crude fibre and neutral detergent fibres. Ca, P, Mg, Zn, Fe, Mn and B were also negatively affected by hand-grabbing and exhibited higher values if the samples were taken by means of the coring device. Crumble losses during manual sampling seem to be a reasonable explanation for these results.

Keywords: sampling methods, core sampler, hand-grabbing, forage quality

Introduction

Forage quality should be periodically ascertained in order to ensure an adequate animal nutrition. Laboratory analyses of forage samples represent the most important tool to collect all the required information. Coring devices allow a standardized sampling of hay (Turnquist *et al.*, 1976; Ball *et al.*, 2001). However, at the present time, hand-grabbing is still the method most frequently used by farmers in South Tyrol to collect hay samples to be analysed. The aim of the present study is elucidating the effect of the sampling method on the results of forage analyses (quality parameters, minerals and trace elements), in order to obtain useful information for their interpretation.

Materials and methods

Hay samples were taken in the autumn of 2013 from piles of loose hay stored in barns at 38 farms in South Tyrol (NE-Italy) distributed over 12 municipalities. Most sampled piles (70%) were made out of field-dried hay, the remaining piles of barn-dried hay. At each farm, piles with hay from the first and from the second cut were sampled. Samples were drawn either by hand-grabbing or by means of a core sampler. Grab samples were obtained from the upper surface of the pile at 15 to 20 evenly distributed, randomly selected spots with a sampling depth up to about 25 cm. The subsamples were pooled and thoroughly mixed. A sample of about 5 l was then taken. Core samples were obtained at three randomly selected spots using a modular, mechanical coring device with a 2.25 cm inner diameter at a sampling depth of 3.5 m. In the lab the samples were ground to pass a 0.5-mm screen using a Fritsch P 25 mill (Fritsch GmbH, Idar-Oberstein, D). Ash was determined according to Naumann *et al.* (1997), crude protein (CP) according to the Dumas method; crude fibre (CF), neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were analysed by means of an Ankom 200 Fiber Analyzer (Ankom Technology, Fairport, USA). The minerals and trace elements were determined according to Naumann *et al.* (1997).

Statistical data analysis was performed with a mixed model taking into account the sampling method and the cut as well as their interaction as fixed effects and the farm as a random effect. The sampling method was considered to be a repeated factor with the sampling event (farm × cut) as a subject. Normal distribution of the residuals and variance homogeneity were consistently checked. Data transformation was performed if necessary. A slight departure from the normal distribution of residuals, which was observed for ADF, Zn and Fe was tolerated. Multiple comparisons were performed by LSD test. $P \leq 0.05$ was considered to be significant.

Results and discussion

The cut was found to affect all quality parameters (Table 1). In accordance with the expectations (Resch *et al.*, 2006), forage obtained with the second cut had higher ash, protein and mineral content and lower fibre content. The sampling method affected eleven of the fifteen investigated parameters: ADF, ADL, K, Na, Cu and Zn were unaffected. The use of the core sampler resulted in higher ash and CP content as well as in lower CF and NDF content. Higher fibre contents of hay samples collected by hand-grabbing in comparison to those obtained by using core samplers have been already observed in sampling trials of alfalfa hay lots (Putnam and Orloff, 2002). Mineral (Ca, P, Mg) and trace (Zn, Fe, Mn, B) elements exhibited lower values if sampled by means of hand-grabbing.

For ash and Fe an interaction between sampling method and cut was detected. In comparison to hand-grabbing, significantly higher ash contents were observed at both cuts if sampling was performed by means of the coring device (94.0 vs 83.8 g kg^{-1} DM at the first cut and 116.7 vs 95.3 g kg^{-1} DM at the second cut), with a larger difference occurring at the second cut. A similar figure was observed for Fe (480.8 vs 734.5 mg kg^{-1} DM at the first cut and 734.7 vs 308.3 mg kg^{-1} DM at the second cut). These differences seem likely to be explained by crumble losses of leafy plant material. On one hand, crumbling

Table 1. Effect of sampling method, cut and their interaction on forage quality parameters (g kg^{-1} DM), minerals (g kg^{-1} DM) and trace elements (mg kg^{-1} DM).

Parameter	Sampling method (SM)		Cut (C)		*P*-values of effects[1]		
	Core sampler	Hand-grabbing	First	Second	SM	C	SM × C
Ash[2]	104.7	89.3	88.7	105.3	**<0.001**	**<0.001**	**0.022**
CP	117.1	111.2	99.0	123.3	**0.010**	**<0.001**	0.601
CF	289.7	301.2	314.8	276.3	**<0.001**	**<0.001**	0.423
NDF	560.7	574.6	602.3	533.1	**<0.001**	**<0.001**	0.749
ADF	364.8	364.6	385.8	343.5	0.909	**<0.001**	0.670
ADL[2]	63.4	61.3	66.3	58.4	0.193	**<0.001**	0.487
Ca[2]	9.17	7.4	7.32	9.30	**<0.001**	**<0.001**	0.749
P	2.87	2.71	2.50	3.08	**0.032**	**<0.001**	0.626
Mg[2]	3.42	2.9	2.73	3.57	**<0.001**	**<0.001**	0.059
K	24.59	24.90	23.13	26.13	0.434	**<0.001**	0.532
Na[2]	0.230	0.223	0.206	0.249	0.629	**0.022**	0.277
Cu[2]	6.70	6.54	5.91	7.42	0.448	**<0.001**	0.203
Zn[2]	39.8	32.9	34.4	38.1	**<0.001**	**0.001**	0.157
Fe[2]	594.4	297.8	371.8	476.0	**<0.001**	**0.005**	**0.012**
Mn[2]	90.6	75.0	77.4	87.8	**<0.001**	**0.028**	0.947
B	16.99	13.54	13.96	16.57	**<0.001**	**<0.001**	0.052

[1] Significant *P*-values are highlighted in bold.

[2] Statistical analysis with logarithm-transformed data. Back-transformed means are shown.

occurring at the surface of the pile during hay handling in the barn may gradually accumulate in the lower part of the pile and be reached only by the core sampler, which allows greater sampling depth than hand-grabbing. On the other hand, losses of crumbled material may occur also during sampling the hay pile by hand-grabbing, mixing the subsamples and taking the 5 l sample to be analysed.

Conclusions

The results provide evidence for a systematic effect of the sampling method on the quality of hay tested by means of laboratory analyses. This effect was observed for the majority of the forage quality parameters, including also mineral and trace elements. Hand-grabbing was found to result in values suggesting lower forage quality than the actual one. It can be assumed that the use of coring devices represents the most reliable way for a standardized and correct sampling of piles of loose hay.

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Effect of dry matter content and feeding level on intake and organic matter digestibility of perennial ryegrass offered to sheep

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Abstract

The objective of this experiment was to evaluate the effect of grass dry matter (DM) content on OMD at two feeding levels in wether sheep. A Latin square experimental design, with four periods and four treatments, was used. There were four sheep per treatment per period (n=16), with the four treatments arranged in a 2×2 factorial; wilted or fresh grass offered at *ad libitum* or maintenance feeding level. Each 12-d period consisted of a 6-d adaptation phase and a 6-d measurement phase (MP). Fresh and wilted grasses were fed to the sheep daily at 4 pm. The DM content of the wilted and fresh grass was 27.8% and 18.6%, respectively. *Ad libitum* fed sheep were offered 110% of the previous day's DMI. Maintenance fed sheep were fed 40 g grass DM kg^{-1} metabolic BW. Sheep were housed in stalls to allow for individual feeding and total faecal collection. During each MP, a sample of the grass fed to, and faeces voided by, each sheep was collected daily. The daily grass and faeces samples were dried and bulked to give one sample per treatment and per sheep per period. Wilted grass (819 g kg^{-1}) had greater OMD than fresh grass (807 g kg^{-1}; $P<0.01$) and maintenance level feeding (825 g kg^{-1}) resulted in greater OMD than *ad libitum* (806 g kg^{-1}; $P<0.001$). When fed *ad libitum*, wilted grass (1.50 kg DM day^{-1}) resulted in greater DMI than fresh grass (1.40 kg DM day^{-1}; $P<0.05$) but fresh grass had greater fresh weight intake (7.5 kg day^{-1}) than wilted grass (5.5 kg day^{-1}, $P<0.001$). These results indicate that increasing perennial ryegrass DM content results in increased OMD and increased DMI in sheep fed indoors, and the results are not interacted upon by the feeding level employed.

Keywords: intake, perennial ryegrass, digestibility, feeding level

Introduction

Grass is a cheap and nutritious feed and Dillon *et al.* (2005) identified grass quality and grass utilisation as key components of profitability in grass-based livestock production systems. Organic matter digestibility (OMD) is a common measurement of grass quality. In order to provide high OMD grass to grazing ruminants it is essential to understand the factors that affect sward quality. High sward OMD is also essential to allow for high voluntary dry matter intake (DMI) of grass by grazing ruminants, although typically in *in vivo* digestibility studies maintenance level feeding produces higher digestibility values than feeding at *ad libitum* levels (Andueza *et al.*, 2013). A major factor affecting DMI is dry matter (DM) content (Cabrera Estrada *et al.*, 2004). The DM content could also have a substantial effect on OMD. However, it is unclear how altering grass DM content would affect digestibility values at both maintenance and *ad libitum* feeding levels. The objective of this experiment was to evaluate the effect of grass dry matter (DM) content on OMD at two feeding levels in wether sheep.

Materials and methods

Sixteen Texel wether sheep were used to evaluate the *in vivo* OMD and voluntary DMI of two grasses differing in DM content at both maintenance and *ad libitum* feeding level. The experiment was run as a 4×4 Latin square design. The four treatments were fresh grass fed *ad libitum*, wilted grass fed *ad* *libitum*, fresh grass fed at maintenance and wilted grass fed at maintenance. There were four periods with 12 d in each period; 6 d adaptation followed by 6 d measurement phase (MP). The experiment was conducted from 27 May to 28 July 2014. In May 2014, four plot areas were established in a sward which was composed predominantly of perennial ryegrass (*Lolium perenne* L.). Each plot was divided into four sub-plots which each provided 3 days feeding. Herbage mass (HM) was measured twice weekly using a Gardena hand shears (Accu 60, Husqvarna AB, Husqvarna, Sweden) and a 0.25 m^2 quadrat (O'Donovan *et al.*, 2002) with a target HM of approximately 1,500 kg DM ha^{-1}. Grass was harvested daily at 08:30 h with a motor lawn mower (Etesia UK Ltd., Warwick, United Kingdom). After harvesting, approximately half the grass was stored at 4 °C and the second half was placed on a 6 m^2 wire mesh platform for wilting. Wilting was achieved using a flow of heated air that passed up through the layer of grass. A centrifugal fan (Sodeca CJBD, Sodeca, Spain) blew air with an outflow of 3,600 m^3 h^{-1} at 18 °C. Grass was dried for 5 hours daily. The sheep were housed individually allowing for the total collection of urine and faeces. Sheep were offered grass twice daily at 15:00 h and at 08:30 h. The grass was offered to the sheep *ad libitum* to allow a 10% refusal rate (previous days grass DMI × 1.1), or at 40 g grass DM per kg metabolic bodyweight for the maintenance fed sheep. Approximately 50% of the grass was offered in the afternoon feeding. The remaining fresh and wilted grass was then refrigerated at 4°C prior to feeding the following morning. The fresh grass offered to each sheep was adjusted daily for DM content by measuring the DM content of the grass. During the MP, a representative sample of the grass offered to, and faeces voided by, each sheep was collected daily. Daily grass and faeces samples were dried and then bulked to give one sample of each per treatment per MP. Data were analysed using a mixed model procedure in SAS. Period, feeding level, grass type and their interactions were fixed effects and sheep was a random effect.

Results

As expected, there was a significant effect of drying grass on grass DM content. Wilted grass had greater DM content than fresh grass (P<0.001) by on average 92 g kg^{-1}. There was no effect of grass DM or feeding level or their interaction on offered grass OM, CP, NDF and ADF concentration (Table 1). As intake for both maintenance feeding level treatments was controlled, both had similar feed intake and were not included when analysing the effect of grass DM on DMI. For the maintenance fed animals DMI was 750±25 g day^{-1}. For *ad libitum* fed animals there was a significant effect of grass DM on DMI (P<0.05). Wilted grass (1.50 kg DM day^{-1}) resulted in greater DMI than fresh grass (1.40 kg DM day^{-1}; P<0.05) but fresh grass (7.5 kg day^{-1}) had greater fresh weight intake than wilted grass (5.5 kg day^{-1}) (P<0.001). There was a significant effect of feeding level on OMD (P<0.05); maintenance level feeding (825 g kg^{-1}) resulted in greater OMD than *ad libitum* (806 g kg^{-1}; P<0.001). There was also a significant effect of DM (P<0.01) on OMD. Wilted grass (819 g kg^{-1}) had greater OMD than fresh grass (807 g kg^{-1}; P<0.01). There was no effect of the feeding level by DM interaction on grass OMD.

Table 1. Effect of dry matter content and feeding level on the chemical composition of perennial ryegrass offered to sheep.[1]

Feeding level	*Ad libitum*		Maintenance		Significance			
DM content	Fresh	Wilted	Fresh	Wilted	s.e.m.	DM	FL	DM*FL
DM (g kg^{-1})	186	278	186	278	1.5	***	ns	ns
OM (g kg^{-1} DM)	870	887	870	884	10.0	ns	ns	ns
CP (g kg^{-1} DM)	213	215	213	213	4.2	ns	ns	ns
NDF (g kg^{-1} DM)	529	513	510	521	10.7	ns	ns	ns
ADF (g kg^{-1} DM)	308	279	275	284	12.9	ns	ns	ns

[1] OM = organic matter, CP = crude protein, NDF = neutral detergent fibre, ADF = acid detergent fibre, s.e.m. = standard error of the mean, ns = P>0.05, *** = P<0.001.

Discussion

The digestibility of grass offered to sheep was high (OMD 813 g kg^{-1}) due to the high proportion of leaf that the swards contained (0.71). In the present study, feeding sheep at maintenance increased OMD by 11 g kg^{-1} and OMD by 19 g kg^{-1} compared to *ad libitum* feeding. This was similar to the findings of (Andueza *et al.* (2013)) who found that at young stages of plant maturity, OMD was higher in maintenance fed sheep than *ad libitum* fed sheep. This could be partially explained by the fact that forage remains in the rumen for a longer period when animals are fed at maintenance level compared to *ad libitum* level (Blaxter *et al.*, 1956). In the present study there was a 12 g kg^{-1} increase in OMD recorded when sheep were offered wilted grass compared to fresh grass. This may also be due a slower rate of passage through the gastrointestinal tract of the sheep. Importantly the effect of the interaction between feeding level and grass DM content on OMD was not significant. This demonstrates that the feeding level employed will not disproportionately affect the effect of grass DM content on grass OMD.

Conclusions

Wilting grass resulted in a greater DMI than fresh grass. There was no effect of the interaction between feeding level and grass DM content on OMD, which demonstrates that the effect of grass DM content can be measured on animals fed at either feeding level. Feeding at maintenance increased OMD compared to *ad libitum* feeding and wilted grass had a higher OMD than fresh grass.

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Cereal-legume mixtures: forage quality under Mediterranean conditions

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Abstract

Forage quality was assessed for pea and vetch-based mixtures with triticale and oat in a Mediterranean environment in 2014 and 2015. Eight binary and two four-component mixtures were analysed and their dry matter, crude protein, fibres, digestibility and ash content determined. Mixtures showed a higher variability in fibre content and digestibility in 2014 than in 2015. The binary mixtures based on common vetch showed higher protein content compared to the other mixtures in both years of cultivation. Pea mixtures showed intermediate protein content between common vetch-based and Narbon vetch-based mixtures. NDF values allowed to classify all mixtures as good (NDF<500 g kg^{-1}). ADF contents in all mixtures were excellent, especially in 2014 when all mixtures showed ADF values below 310 g kg^{-1}. DDM was quite good in all mixtures ranging from 597 to 678 g kg^{-1}. Triticale showed a marked positive effect on the forage quality of binary mixtures regardless of the companion legume, improving CP and DDM and reducing fibre contents. These preliminary results allowed us to identify some winter cereal-legume mixtures with high forage quality.

Keywords: pea-based mixtures, vetch-based mixtures, oat, triticale, forage quality

Introduction

Winter annual forage crops represent one of the key components of most Mediterranean farming systems. Mixtures of annual forage legumes with winter cereals for forage production are used traditionally in the Mediterranean region. However, the self-sufficiency in the production of high protein content feed in Mediterranean farming systems is a challenge especially for organic systems and with animals having a high demand of proteins for an optimal growth and production. In this framework, the adoption of new cereal-legume mixtures into the forage systems might satisfy at the same time the protein and the energy requirements of animals and a higher economic profitability for farmers. In order to face this challenge, the European project REFORMA aimed at developing new varieties of legumes and at promoting existing varieties to be used in optimal diets produced 'on farm'. The new varieties include innovative high-yielding pea forage crops with a greater drought tolerance and lodging resistance than the available varieties for Mediterranean areas. This paper reports the results of a comparison between pea-cereal-based and vetch-cereal-based mixtures focusing on some forage quality traits.

Materials and methods

The experiment was carried out in the years 2013-2014 and 2014-2015 in the experimental field of CNR-ISPAAM (40° 45' N, 8° 25' E, 24 m a.s.l.). The site is characterized by a typical central Mediterranean climate (550 mm of average annual rainfall) and deep alluvial calcareous soil (pH=7.8). Four legumes and two winter cereals were used to compose binary and four-component mixtures: *Pisum sativum* L., semi-leafless semi-tall type 'Kaspa' (P1) and semi-leafless tall type 'line 2/37b' (P2) (from CREA, Lodi – Italy), forage pea; *Vicia narbonensis* L. 'Bozdag'(N), Narbon vetch; *Vicia sativa* L. 'Barril' (V), common vetch; *Avena sativa* L. 'Genziana' (O), oat, and x*Triticosecale* 'Amarillo' (T), triticale. The species were mixed in binary mixtures using 35 viable seeds m^{-2} for P1, P2 and N, 70 seeds m^{-2} for V, 140 seeds m^{-2} for O and T. The number of seeds m^{-2} was halved for each species in four-component mixtures. A total of ten mixtures, eight binary mixtures (P1O, P1T, P2O, P2T, NO, NT, VO, VT) and two four-component mixtures

(P1P2OT and NVOT) were grown in plots (4×3 m) following a complete randomized block design with 4 replications. Before sowing, nitrogen and phosphate fertilizers were distributed (15 kg nitrogen and 45 kg P_2O_5 ha^{-1}). The same amount of nitrogen was distributed in each plot at the end of the winter. Sowing was done on 7th November 2013 and 21 November 2014. Harvest of biomass for hay production was done in May in both years. The harvest time was chosen on the basis of legume phenological stage (early pod-filling) and cereals at heading stage. Herbage samples were taken from a quadrat (1 m^2) in the centre of each plot. A subsample was dried in ventilated oven at 60 °C up to constant weight. Dry matter (DM) was then ground and analysed for crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre (NDF) and ash contents (Goering and Van Soest, 1970) and for digestible dry matter (DDM) measured as an enzymatic solubility (Lila *et al.*, 1986). Two repetitions were analysed. One way ANOVA was performed with Statgraphics Centurion XV (Statpoint Technologies, Inc. Warrenton, Virginia). When means were statistically different, LSD values (P<0.05) were calculated.

Results and discussion

Mixtures quality varied between years 2014 and 2015 (Table 1). In general terms, all observed parameters differed among accessions most in 2014 than in 2015, with the exception of ash content, which values were statistically similar in both years. CP and DDM were higher in 2014 than in 2015 (113 and 636 g kg^{-1} vs 95 and 616 g kg^{-1}, respectively). ADF and NDF were significantly higher in 2015 than in 2014 (306 and 495 g kg^{-1} vs 284 and 466 g kg^{-1}, respectively). Finally, DM was higher in second year (236 vs 186 g kg^{-1}). These results can be partially explained by the different meteorological trend between the two years. During the first growing season (Oct 2013-May 2014), the total amount of rainfall (529 mm) was close to the plurennial average while the following growing season (Oct 2014-May 2015) was quite dry (440 mm). Yet, temperatures were slightly more favourable to mixtures growth in the winter of the first growing season. A comparison of the quality of the mixtures used in this trial with the one of others used in other experiments is quite complicated mainly due to the different seed ratios and cereal species/cultivars used. Nonetheless, some general considerations can be done referring to general forage quality standards for livestock diets. A forage having CP equal to 120-130 g kg^{-1} is considered to be of good quality and considered sufficient to meet most protein sheep and beef cattle requirements (except for some categories with higher requirements, i.e. ewes suckling twins) (Haj-Ayed *et al.*, 2000). From this point of view, V-based mixtures were of good quality (VO, NVOT) or excellent (156 g kg^{-1} in VT), and P1-P2- and N-based mixtures were of medium quality. Binary mixtures based on V showed a higher CP respect to the other mixtures in both years of cultivation. P1/P2 mixtures showed an intermediate protein content between V-based and N-based mixtures, ranging from 86 and 110 g kg^{-1}. N-mixtures showed the lowest protein content, especially in 2015.

Mixtures differed for NDF, ranging from 434 (P2T in 2014) to 525 g kg^{-1} (NT in 2015). Despite this, NDF values allowed us to classify all mixtures as good (NDF<500 g kg^{-1}). Significant differences for ADF values were found among mixtures only in the first year, ranging from 266 (NT) to 304 g kg^{-1} (VT). Likewise, ADF contents in all mixtures were excellent, especially in 2014 when all mixtures showed ADF rates below 310 g kg^{-1}. In 2015, some mixtures (P1T, P2O, and NVOT mixtures) exceeded this maximum desirable value. Nonetheless, their ADF percentage was still acceptable. DDM was quite good in all mixtures ranging from 597 (P2O in 2015) and 678 g kg^{-1} (NT in 2014). No significant differences were found among mixtures for DDM in the second year. As a whole, triticale showed a marked positive effect on the forage quality of binary mixtures regardless of the companion legume, improving CP and DDM and reducing fibre contents.

Table 1. CP, NDF, ADF, DDM and ashes content of binary and 4-component mixtures based on peas (P1, P2), vetches (V, N), oat (O) and triticale (T). LSD values were calculated for *P*<5%.

Mixture	Legume (%)		CP (g kg^{-1})		NDF (g kg^{-1})		ADF (g kg^{-1})		DDM (g kg^{-1})		Ashes (g kg^{-1})	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
NO	19.8	18.0	90	69	512	512	284	290	617	614	100	85
NT	23.9	23.3	105	69	486	525	266	307	678	599	85	74
P1O	60.0	41.5	88	86	461	480	284	295	627	626	78	87
P1T	68.3	48.8	107	92	438	500	273	318	662	609	73	79
P2O	59.5	34.8	110	83	476	516	297	314	600	597	90	87
P2T	83.2	59.8	104	99	434	467	278	301	647	622	62	80
VO	59.1	38.5	131	113	482	503	296	308	615	622	99	104
VT	80.3	68.0	156	144	464	457	304	307	636	640	108	104
NVOT	56.5	53.3	135	110	465	492	282	314	644	627	105	104
P1P2OT	69.3	35.3	109	85	446	503	280	309	636	600	78	88
LSD	16.2	20.3	19.6	18.9	45.6	66.7	26.6	33.9	46.9	54.3	15.2	8.1

Conclusions

These preliminary results allowed us to identify some winter cereal-legume mixtures with high forage quality. Nonetheless, the forage quality offered on a hectare basis requires the collection of DMY data to assess the actual value of mixtures.

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Nitrogen fertilisation effects on multi-species and *Lolium perenne* yields and composition under silage management

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Abstract

This study examined the effects of nitrogen (N) fertilisation on the yield and chemical composition of multi-species and *Lolium perenne* swards under a four-cut silage management regime. Multi-species swards produced greater annual yields than *L. perenne* and increasing N application rate increased yield. This latter effect did not interact with species for the first or final cuts, but *L. perenne* had a greater yield response to N fertilisation than the multi-species swards for both mid-season cuts. Herbage nutritive value was not affected by species, however N application increased crude protein concentration. Ensilability indices indicated that *L. perenne* was likely to preserve satisfactorily as silage with greater ease than multi-species swards, however increasing N application reduced herbage ensilability similarly for both sward types.

Keywords: multi-species swards, nitrogen, silage, yield, chemical composition

Introduction

In temperate north-western Europe, *Lolium perenne* is the primary grass sown for pasture-based ruminant production. These swards perform well under both grazing and cutting regimes where soil fertility is adequate and inputs of inorganic nitrogen (N) are relatively high. Research suggests that, compared to *Lolium perenne* monocultures, multi-species swards can increase herbage yields at similarly high N fertiliser inputs or even maintain herbage yields at reduced N fertiliser input (Nyfeler *et al.*, 2009; Connolly *et al.*, 2009). These benefits have been attributed to positive interspecific interactions and increased nutrient-use efficiency as functional diversity increases (Hector *et al.*, 1999). Less is known, however, of the nutritive value and ensilability of multi-species swards. Thus, the objective of this study was to quantify the effects of a series of N fertilisation rates on the dry matter (DM) yield and chemical composition (primary growth only) of two multi-species swards relative to a *L. perenne* monoculture, under a four-cut silage management regime.

Materials and methods

Field plots (each 10×2 m) were organised in a randomised complete block (n=4) design and had four successive cuts taken throughout the growing season: 27 May, 15 July, 2 September and 11 November 2014. Each block contained 12 plots to which were allocated three herbage species receiving four rates of inorganic N fertiliser. Species treatments were: *L. perenne* monoculture (*Lp*), Mix 1 (*L. perenne, Phleum pratense, Trifolium pratense* and *repens*) and Mix 2 (*L. perenne, P. pratense, T. pratense, Cichorium intybus* and *Plantago lanceolata*). The four rates of inorganic N fertiliser, applied as calcium ammonium nitrate (275 g N kg^{-1}) were 0, 120, 240 and 360 kg N ha^{-1} $annum^{-1}$, with 33, 28, 22 and 16% of the total application allocated to the herbage managed for Cuts 1, 2, 3 and 4, respectively. Herbage was cut to a stubble height of approximately 5.5 cm using a Haldrup plot harvester. Yield and chemical composition were determined as reported by King *et al.* (2012). Statistical analysis was undertaken using a general linear model which accounted for species, rate of N, species × rate of N, block and residual effects. The rate of N application effects were separated into their linear, quadratic and cubic components.

Results and discussion

The interactions between species and rate of N were not significant (P>0.05) unless otherwise stated. Although species did not affect (P>0.05) the yield of Cut 1, for each of Cuts 2, 3 and 4 both Mix 1 and Mix 2 yielded greater (P<0.001) than *L. perenne* and there was no difference between the two mixed-species treatments (Table 1). Overall, there was a similar annual production advantage to both multi-species swards compared to *L. perenne* (13,539 versus 11,891 kg DM ha^{-1}). Annual yields were relatively high even when no inorganic N was applied, and for each cut there was a linear increase in herbage yield in response to N fertiliser application. However, for Cuts 2 and 3 there was a species × rate of N interaction where the yield of *L. perenne* at the two lower N rates was lower (P<0.01) than for the two multi-species swards.

There was no effect (P>0.05) of species on herbage nutritive value traits such as DM digestibility (DMD) and crude protein (CP). While increasing rates of N application did not alter DMD (P>0.05), they did cause a linear increase in CP (P<0.05) and this is in agreement with Conaghan *et al.* (2012). Mixes 1 and 2 would likely be more difficult to satisfactorily preserve as silage than *L. perenne* due to their lower WSC_{Aq} and higher buffering capacity. Although increasing N fertiliser application did not reduce WSC_{Aq} (P>0.05), in disagreement with Keating and O'Kiely (2000), similarly to O'Kiely *et al.* (1997) it did lead to a linear increase in buffering capacity (P<0.05). This suggests that increasing rates of N fertiliser application would make the herbage more difficult to satisfactorily preserve as silage.

Table 1. Herbage yield (kg DM ha^{-1}) per cut and annually, and Cut 1 chemical composition 2014. [1]

	Species					N fertilisation rate (kg ha^{-1} annum^{-1})					
	Lp	Mix 1	Mix 2	SEM	*P*	0	120	240	360	SEM	*P*[2]
Yield											
Cut 1	7,114	6,846	7,095	265.3	0.731	6,429	7,074	7,113	7,458	306.4	0.029
Cut 2	1,998	2,505	2,690	76.1	<0.001	1,659	2,162	2,941	2,830	87.8	0.001
Cut 3	1,585	2,413	2,472	66.7	<0.001	1,736	1,906	2,410	2,574	77.1	0.001
Cut 4	1,193	1,463	1,596	57.6	<0.001	1,089	1,406	1,649	1,524	66.5	0.001
Annual	11,891	13,227	13,852	256.5	<0.001	10,913	12,547	14,113	14,386	296.2	0.001
DM	154	138	142	3.6	0.01	152	145	138	143	4.2	0.082
DMD	682	695	684	6.1	0.279	698	685	682	682	7.1	0.103
CP	101	100	106	3.9	0.477	95	102	105	108	4.5	0.041
WSC_{DM}	133	107	101	2	0.05	131	113	101	109	2.3	0.086
WSC_{Aq}	25	17	17	9	0.05	24	19	17	19	10.3	0.107
BC	274	293	296	5.5	0.05	278	283	293	295	6.4	0.036

[1] *Lp* = *Lolium perenne*; SEM = standard error of mean; DM = dry matter (g kg^{-1}); DMD = DM digestibility (g kg^{-1}); CP = crude protein (g kg^{-1} DM); WSC = water-soluble carbohydrates (WSC_{DM}: g kg^{-1} DM; WSC_{Aq}: g l^{-1}); BC = buffering capacity (mEq kg^{-1} DM). Cut 2 yields were 814, 1691, 2,842, 2,646; 1,979, 2,248, 2,928, 2,866; 2,183, 2,546, 3,053 and 2,977 (SEM 152.1; P<0.01) kg DM ha^{-1} and Cut 3 yields were 566, 1,376, 2,060, 2,339; 2,296, 2,209, 2,445, 2,703; 2,346, 2,134, 2,727 and 2,681 (SEM 133.1; P<0.001) kg DM ha^{-1}, for *L. perenne*,, Mix 1 and Mix 2, each at 0, 120, 240 and 360 kg N ha^{-1} annum^{-1}, respectively.

[2] *P*-value of linear component.

Conclusions

Overall, both multi-species swards produced similar annual yields and these were greater than yields achieved with *L. perenne*. Although the yield response to N fertiliser was greater for *L. perenne* than the multi-species mixtures for Cuts 2 and 3, it did not differ for the other cuts or for annual yield. For Cut 1, neither the absence of a species effect on herbage nutritive value nor the more challenging ensilability characteristics of the multi-species mixtures than *L. perenne* interacted with the rate of fertiliser N applied.

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Widening the harvest window with contrasting grass-clover mixtures

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Abstract

Widening the harvest window for the first silage cut is crucial in adapting to unstable weather conditions without degrading the silage nutrient content. This study investigated whether a grass-clover mixture containing late species and varieties harvested late could provide the same yield and nutritional quality as a mixture with early species and varieties harvested early. Trials based on a split-plot design (four replicates), with harvest time on main plots and seed mixture on sub-plots, were conducted at three sites in Sweden in 2007-2009. The results showed that on average for all three sites and ley years, there were no significant differences in silage yield, energy content or fibre content between the early mixture harvested early and the late mixture harvested late. Thus in practice the harvest window can be widened by 9 days for the first cut by using an early cut mixture containing early varieties of timothy, meadow fescue, perennial ryegrass and clover, or a late cut mixture containing late varieties of timothy, perennial ryegrass and clover, but without meadow fescue.

Keywords: seed mixture, harvest time, forage, yield, energy, NDF

Introduction

It is important to harvest silage leys at the optimal time to obtain high-quality forage for dairy cows and growing cattle. Harvesting strategy is important for yield and quality, e.g. different forage species have different heading dates, affecting their optimal harvest time (Halling, 2012). Within species, there are differences in earliness depending on type, e.g. timothy (*Phleum pratense* L.) SW Ragnar is generally three days later than Grindstad (Halling, 2012). One way to widen the harvest window is to choose a mixture of species and varieties which together provide a slower decrease in energy over time. Another is to sow different mixtures that have their optimum harvest period at different times. This project investigated whether a seed mixture containing late species and varieties harvested late can give equivalent yield and nutritional quality to a mixture with early species and varieties harvested early.

Materials and methods

Trials based on a split-plot design with four replicates, with harvest time on main plots and seed mixture on sub-plots, were conducted in Jönköping (57°44'00.5'N; 14°16'44.0'E), Kalmar (56°41'29.0'N; 16°13'16.7'E) and Rådde (57°36'25.1'N; 13°15'25.8'E), Sweden, over a three-year period (2007-2009) (Table 1). The leys were undersown (seed rate 20 kg ha^{-1}) in cereals in 2006 at all sites except Jönköping, where no cover crop was present. The sward was harvested three times per year. For comparison with late and early mixtures, a standard mixture containing 15% timothy (TI) cv. Grindstad, 30% TI Ragnar, 20% meadow fescue (MF) (*Festuca pratensis* Huds.), Sigmund and Tyko, 20% perennial ryegrass (PR) (*Lolium perenne* L.) Helmer, 10% red clover (RC) (*Trifolium pratense* L.) Sara and 5% white clover (WC) (*Trifolium repens* L.) Ramona was sown. The early grass-clover mixture contained 25% TI Grindstad, 40% MF, Sigmund and Tyko, 20% PR, Gunne and Baristra, 10% RC Titus and 5% WC Ramona. The early grass mixture included 30% TI, 45% MF and 25% PR (same varieties); the late grass-clover mixture 40% TI Ragnar and Comtal, 45% PR Herbie and Tivoli, 10% RC Vivi and 5% WC Ramona; and the late grass mixture 50% TI and 50% PR (same varieties). In each growing season, a total of 140 kg N ha^{-1} was applied to the grass-clover mixtures (55 + 45 + 40 kg N ha^{-1} divided between cuts) and 100

Table 1. Experimental design with harvest time in main plots and seed mixture in sub-plots.

Harvest time	Seed mixture
S1. First harvest at approx. 11 MJ in mixture D	A. Standard mixture
S2. First harvest at approx. 11 MJ in mixture E or 10-12 days after S1	B. Early grass-clover mixture
	C. Late grass-clover mixture
	D. Early grass mixture
	E. Late grass mixture

+ 80 + 60 kg N ha^{-1}; 240 N ha^{-1} to the grass mixtures. The chemical composition of the mixtures was determined at each harvest using standard wet chemistry methods described by Eriksson *et al.* (2004) for dry matter (DM) (at 103 °C), crude protein (6.25 × Kjeldahl N) and ash-free NDF with amylase and sodium sulphite treatment. Metabolisable energy (ME) was calculated from 96-h *in vitro* organic matter digestibility with the VOS method (Volden, 2011), using the grass equation. Statistical analysis was performed using the Mixed model procedure in SAS Version 9.3 (SAS Institute Inc., 2010). Fixed factors were treatment factors, ley year and site.

Results and discussion

Statistical analysis (ley year × seed mixture × harvest time) showed significantly different ($P<0.058$) yield at first harvest, but BS1-CS2 and DS1-ES2 did not differ significantly in terms of first-year ley yield, or BS1-CS2 in terms of third-year yield. However, there were significant differences in second-year yield, with the late harvested late mixtures producing approx. 600-700 kg ha^{-1} more than the early mixtures harvested early. The PR was only affected by moderate winter losses in the third winter, but the ground cover in the following spring in the late mixtures with 45-50% sown PR was approx. 10% units lower than in the early mixtures. On average for all three sites and ley years, there were no significant differences in energy or fibre content between the early mixtures harvested early and the late mixtures harvested late.

For *total* DM yield as an average over all three ley years, there were interactions between harvest time, mixture and ley year ($P<0.050$) and site ($P<0.041$). There were no differences between the early mixtures harvested early and late mixtures harvested late except in Kalmar, where the early grass-clover mixture harvested early yielded approx. 1150 kg DM more per ha than the late, probably due to dry weather between S1 and S2 (Table 2). As regards the nutritional quality of three ley years, CS2 did not differ from BS1 in energy content (harvest time × seed mixture; $P<0.001$) (Table 3). In contrast, the proportion of fibre was 25 g NDF kg^{-1} DM higher in BS1 than in CS2 (harvest time × seed mixture; $P<0.011$).

It is possible to widen the harvest window only by using late timothy and late red clover with no perennial ryegrass. However, it is much easier to use an early mixture including early varieties of timothy and meadow/tall fescue (*F. arundinacea* Schreb.)/*Festulolium*. Another way to widen the harvest window is to use later varieties in one mixture and even earlier varieties in another. With 3-4 days between the harvest time of two mixtures and winter-tolerant species, the harvest strategy could be coordinated with other grassland farms. Use of both late and early seed mixtures enables a late mixture harvested early to be used as 'power feed' combined with protein-rich regrowth in the feed ration. However, considering weaknesses in overwintering capacity, a large proportion of perennial ryegrass in the sward is a high-risk choice at high latitudes.

Conclusions

The harvest window can be widened by 9 days for the first cut by using an early cut mixture containing early varieties of timothy, meadow fescue, perennial ryegrass and clover, or a late cut mixture containing late varieties of timothy, perennial ryegrass and clover, but without meadow fescue.

Table 2. DM yield (kg ha^{-1}) and relative yield of seed mixtures A-E (with/without legumes) at harvest times S1 and S2. Total of three harvests at three sites, mean of ley years.

Seed mixture	Harvest time	Jönköping Yield kg	Jönköping Rel. Mix.	Jönköping Rel. Time	Kalmar Yield kg	Kalmar Rel. Mix.	Kalmar Rel. Time	Rådde Yield kg	Rådde Rel. Mix.	Rådde Rel. Time
A. Standard	S1	13,290^{cde}	100	100	12,390^{hij}	100	100	11,270^{no}	100	100
B. Early – leg.	S1	14,120^{ab}	106	100	12,500^{ghi}	101	100	11,560^{lmn}	103	100
C. Late – leg.	S1	12,240^{hijkl}	92	100	11,860^{klmn}	96	100	10,700^{p}	95	100
D. Early – no leg.	S1	nd			11,980^{ijklm}	97	100	11,870^{ijklm,}	105	100
E. Late – no leg.	S1	nd			11,570^{lmn}	93	100	10,800^{op}	96	100
A. Standard	S2	13,950^{abc}	100	105	11,730^{jlmn}	100	95	12,710^{efgh}	100	113
B. Early – leg.	S2	14,230^{a}	102	101	12,130^{hijkl}	103	97	13,090^{deg}	103	113
C. Late – leg.	S2	13,470^{bcd}	97	110	11,350^{mnop}	97	96	11,570^{lmn}	91	108
D. Early – no leg.	S2	13,260^{df}	95		12,390^{hik}	106	103	13,460^{bcd}	106	113
E. Late – no leg.	S2	12,720^{egh}	91		11,430^{mno}	97	99	12,010^{ijklm}	94	111

P-value (seed mixture × harvest time × site) = 0.041.

$^{a-p}$ Values with different letters are significantly different ($P<0.05$), nd = not determined.

Table 3. DM yield (ha^{-1}), and crude protein, energy and fibre content (kg^{-1} DM) in grass-clover seed mixtures A-C at harvest times S1 and S2. Total of three harvests, mean of three sites and three ley years.

Variable	A – standard		B – early		C – late		*P*-value
	S1	S2	S1	S2	S1	S2	
Yield, kg DM	12320	12800	12730	13150	11600	12130	ns
Crude protein, g	161	143	153	137	168	153	ns
Energy, MJ	10.5^{b}	10.3^{c}	10.4^{b}	10.2^{d}	10.7^{a}	10.5^{b}	$P<0.001$
Fibre, g NDF	475^{d}	506^{b}	495^{c}	521^{a}	454^{e}	470^{d}	$P<0.011$

$^{a-e}$ Values within rows with different letters are significantly different ($P<0.05$), ns = non-significant.

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The potential of α-linolenic acid to predict herbage quality

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Abstract

The calculation of the energy value of herbage relies on the digestibility of the organic matter which in turn is estimated by regression from proximate analysis with crude protein and crude fibre or ADF being the most important predictors. It has been shown in pure swards that particularly α-linolenic acid evolves with the phenological stage which conveys a predictive potential to this fatty acid. That assumption was tested for mixed swards. Fatty acid profiles were analysed in 76 herbage samples from 14 dairy farms situated in contrasting climatic zones in Switzerland to document changes in the nutrient and fatty acid contents over the spring growth period. Fibre content increased from 120 to 300 g kg^{-1} DM and protein content decreased from 220 to below 100 g kg^{-1} DM. The α-linolenic acid proved to be the most dominant fatty acid accounting on average for 50% in the extracted fat. High correlations were found between α-linolenic acid (g g^{-1} extracted fat) and crude fibre content (g kg^{-1} DM) (r=-0.71), ADF (r=-0.69), crude protein (r=0.79) and extracted fat (r=0.79). These relations confirm that α-linolenic acid evolves with the nutritive value and has a predictive potential for mixed swards.

Keywords: α-linolenic acid, proximate analysis, herbage quality, spring growth

Introduction

The calculation of the energy value of herbage relies on the digestibility of the organic matter (dOM) which is either determined *in vivo*, *in vitro* or estimated by regression from proximate analysis. The latter approach is widely used with crude protein (CP) and crude fibre (CF) or ADF being the most important predictors. The regressions applied in Switzerland for mixed swards differentiate between 7 botanical types and include correction factors for cycle number and growth stage. The prediction accuracy is limited in cases of unknown botanical composition and growth stage. In addition to the classical feed nutrients, more specific predictors relating to functional plant traits that vary with plant maturity have the potential to improve the prediction accuracy in a generalized manner. In this context it has been shown that particularly α-linolenic acid (C18:3n-3) evolves with the phenological stages and varies with plant species and plant parts (Boufaïed *et al.*, 2003; Wyss and Collomb, 2010; Wyss, 2012). C18:3n-3 is present in high proportions in the thylakoid membranes of chloroplasts (Hawke, 1973) where it regulates the membrane fluidity of the photosynthetic tissues, i.e. the leaves. Thus, variations in C18:3n-3 reflect variations in leaf/stem ratio, leaf age and temperature and relate C18:3n-3 to herbage quality. In our study, that assumption was tested for mixed swards. This perspective completes most studies investigating first of all links between fatty acids in herbage and milk (Khiaosa-ard *et al.*, 2015).

Materials and methods

During the spring period of 2014, herbage growth and quality were monitored from mid-march to mid-June in experimental plots of 14 farms situated between 450 and 1200 m a.s.l. in contrasting climate zones of western Switzerland. Experimental plots represented the typical wide range of meadow and pasture types used in dairy cow feeding ranging from grass-legume leys to evolved old leys and permanent grassland with varying productivity level and botanical composition. Proximate analysis was carried out throughout the spring period while amino acid and fatty acid profiles were analysed in a reduced sample set at intervals corresponding to early, mid, and late growth stages. Fatty acid profiles were obtained by an improved fat extraction method based on gas chromatography (GC) with in situ transesterification and solid-phase-extraction (Ampuero Kragten *et al.*, 2014). FatGC refers to the sum of fatty acids calculated as triglycerides. Results of the most important fatty acids are given in absolute and relative quantities. Data analysis emphasized on evolution patterns and correlations.

Results and discussion

Phenological changes occurring during spring growth are associated with decreasing protein and fat content and increasing cell wall constituents (CF, ADF, NDF) which reduce the digestibility of the organic matter (dOM) as observed in the present study (Table 1). Fibre content increased from 120 to 300 g kg^{-1} DM and protein content decreased from 220 to below 100 g kg^{-1} DM. As expected, α-linolenic acid proved to be the most dominant and most variable fatty acid, on average accounting for 50% of the extracted fat (Table 1). Figure 1 illustrates the transient reduction in absolute and relative C18:3n-3 content over the spring growth period dropping from 25 to 4 g kg^{-1} DM (Figure 1A) and from 60 to 40% of extracted fat (Figure 1B), respectively. This declining pattern was observed across all sites giving strong evidence for the relationship that exists between C18:3n3 and plant phenology (maturity) and herbage quality at harvest. The level of C18:3n-3 content gives an additional indication for sward use intensity. The two extensively used sites 'Moudon ext' and 'Puidoux ext', both having high proportion of herbs and non-ryegrass gramineae, figure at the low end of the range. Figure 1 also reveals the altitudinal gradient with a temporal shift (sites >900 m: St-George, La St-George and La Frêtaz).

Relating C18:3n-3 content and proportion with nutrient contents of herbage resulted in high positive correlations to CP and fat content and negative correlations to fibre fractions and dOM (Table 1) which confirms previous results of investigations with grass and maize silages (Khan *et al.,* 2012). Another interesting outcome of the present study is the high positive correlation between C18:3n-3 and the

Table 1. Proximate analysis, calculated digestibility, fatty acids and correlations between C18:3n-3 and nutrients of herbage from 14 sites sampled over the spring period.

Nutrients	Overall	Early growth stage	Late growth stage	Correlation C18:3n-3 g kg^{-1} DM	Correlation C18:3n-3 g g^{-1} fat (GC)
Complete data set (76)	**avg**	**avg**	**avg**	**r**	**r**
Ash g kg^{-1} DM	90.6	100.7	73.8		
CP g kg^{-1} DM	168.1	198.9	115.8	0.933	0.786
Cfat g kg^{-1} DM	35.8	41.0	26.7		0.672
CF g kg^{-1} DM	190.0	151.3	259.8	-0.880	-0.709
ADF g kg^{-1} DM	220.6	179.9	294.1	-0.859	-0.693
NDF g kg^{-1} DM	385.2	327.8	487.5	-0.823	-0.659
dOM %	77.9	80.7	71.4	0.774	0.647
Reduced data set (41)					
FatGC g kg^{-1} DM	29.4	37.3	21.6		0.786
C16:0 g kg^{-1} DM	4.11	4.90	3.27		
C18:2n-6 g kg^{-1} DM	4.97	5.85	4.02		
C18:3n-3 g kg^{-1} DM	14.78	19.82	9.98		
C16:0 g g^{-1} fatGC	0.14	0.13	0.15		
C18:2n-6 g g^{-1} fatGC	0.17	0.16	0.19		
C18:3n-3 g g^{-1} fatGC	0.49	0.53	0.46		
Tyr g 100 g^{-1} CP	3.19	3.43	2.94	0.857	0.741

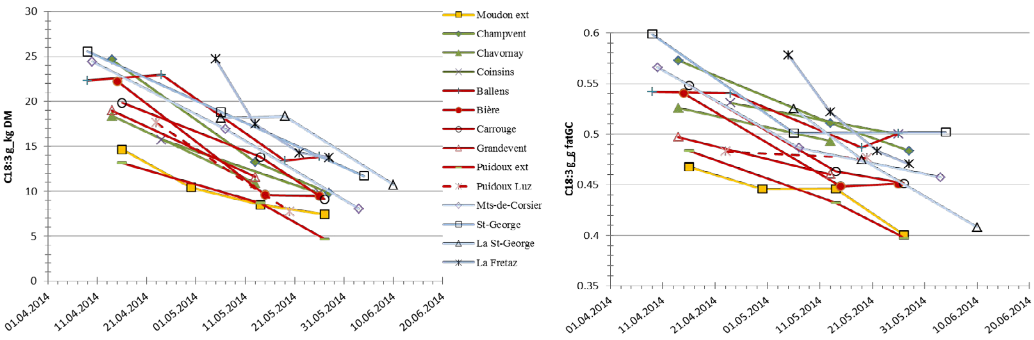

Figure 1. Evolution of C18:3n-3 in herbage from 14 sites over the spring period: (A) in g kg^{-1} DM; (B) g g^{-1} fat (GC).

amino acid tyrosine (Table 1). Given the function of tyrosine as an electron carrier in the photosystem II, that finding is not surprising after all.

Conclusions

As underlined by our results, C18:3n-3 content and proportion decline with advancing maturity of mixed swards and correlate with protein content, cell wall components and calculated digestibility. The involvement of C18:3n-3 in the photosynthetic process of the highly digestible leaves conveys to α-linolenic acid the potential to improve the accuracy of prediction equations that estimate the nutritive value of herbage. It can be assumed that the same holds true for tyrosine. Next steps should analyse the behaviour of C18:3n-3 in herbage of regrowth cycles and evaluate in a series of digestibility trials the relative contribution of α-linolenic acid to the prediction accuracy of multiple regressions. In case of a relevant contribution, a NIR calibration of the expensive fatty acid analysis may be worth the effort.

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Use of near infrared reflectance spectroscopy for the determination of silica content in tall fescue

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Abstract

Major disadvantages of using tall fescue as forage grass are its low digestibility and animal voluntary intake. One of the factors explaining this is the high silica content of tall fescue in comparison with other forage grass species. Therefore, we evaluated the possibility to use NIRS on grass samples to predict the silica content. In 2014, we collected 297 samples on four different trials with tall fescue. These samples were analysed by wet chemistry for silica content, the NIRS spectra were recorded and a calibration model was built. In 2015, forage samples of 25 genotypes in a breeding nursery were collected and used for validation of the calibration equation. The mean silica content in this sample was 0.56% with a standard deviation of 0.14%. The validation statistics indicated that prediction of the silica content for this independent validation sample was good (SEP 0.063%; R^2= 82.2%). The large variation in silica content found within the tall fescue genotypes and the ease of prediction using NIRS are a good basis for selecting tall fescue varieties with lower silica content.

Keywords: *Festuca arundinacea,* breeding, animal preference, silicon

Introduction

There is no consensus whether silicon – commonly referred to as silica in ecological research because of its hardly separable bound with two oxygen molecules – is essential or not to plants. It is however certain that silica plays an important role in the tolerance against different kinds of stress (Guntzer *et al.*, 2012). In grass species e.g. silica protects against herbivory: in sheep preference trials, sheep grazed longer, took more bites and had a higher bite rate on grass species with low silica content (Massey *et al.*, 2009).Tall fescue (*Festuca arundinacea* Schreb.) is a forage grass species with interesting properties in the light of climate change such as good drought resistance and high yield potential (Cougnon et al, 2014). Its low voluntary intake and low digestibility however, are damping the use of the species by farmers, and are brought into relation with the high silica content of this species compared to other forage grass species like perennial ryegrass (*Lolium perenne*) and meadow fescue (*Festuca pratensis*) (Hodson *et al.*, 2005). Hence, studying the silica content of tall fescue genotypes can help to elucidate the factors that influence the low animal preference of this species and can eventually lead to varieties with an improved animal preference.

The screening of a large amount of genotypes is hampered by the cumbersome analytical method. near infrared reflectance spectroscopy (NIRS) is a high throughput method that is often used in grassland science to predict a wide range of parameters like crude protein content, digestibility or clover content of swards (Cougnon *et al.*, 2013). Smis *et al.* (2014) developed a NIRS calibration equation that allowed testing the Si content of different forest plant species. We tested whether it was possible to develop a NIRS calibration for silica content specific for tall fescue.

Materials and methods

Both the NIRS spectra and the Si content of 595 dried and ground grass samples from different trials were determined during the years 2014 and 2015. Silica content was analysed using the wet chemistry method described by the alkaline method of DeMaster (1981). In brief, about 25-30 mg grinded and homogenized plant material was mixed with 25 ml of Na_2CO_3 solution (0.1 M) and incubated in a water bath maintained at 85 °C for 4 hours. After filtration (0.45 µm), the extractions were colorimetrically analysed for Si. NIRS spectra were recorded on a FOSS XDS Rapid Content Analyzer The inverse reflectance [log(1/R)] was measured from 400 to 2,500 nm in steps of 0.5 nm. From the 595 samples, 165 were collected on a trial comparing the sheep preference of 19 genotypes of tall fescue on three occasions in 2014; 57 were collected from plants in a tall fescue clonal nursery on three occasions in 2014; 6 were collected in a tall fescue-perennial ryegrass pasture on one occasion in 2014; 69 samples originated from a trail were the agronomic performance of different varieties of tall fescue, *Festulolium*, meadow fescue and perennial ryegrass was compared in 5 cuts in 2014; finally 298 samples were harvested on two occasions in 2015 in a study of 25 clones and their progeny in three replicates.

The 297 samples collected in 2014 were used to build a calibration equation. The 298 samples harvested in 2015 were used to validate the equation. The validation samples were independent from the calibration samples as they were harvested in a different year and on a different trial than the calibration samples. Calibration equations were developed in WinISI II 1.5 (Infrasoft, Port Mathilda, PA, USA) using the partial least square regression method. A data pre-treatment was performed before regression: the first derivative of the spectra was taken (with gap 4 and smoothing 4) and scatter correction using standard normal variate (SNV) and detrend was applied. Standard errors of calibration, cross validation and prediction (SEC, SECV and SEP) were calculated, and the R^2 of the regression between the lab values and the predicted values was calculated. No outliers were removed from the calibration or the validation sets.

Results and discussion

The silica content in the 297 calibration ranged from 0.21 to 1.46%, with an average content of 0.62% and standard deviation of 0.19%. The mean Si content in the validation set was 0.56% with standard deviation 0.14%. An equation with 15 variables was selected by minimizing the SECV: the SEC and SECV for this equation were 0.056 and 0.067% the R^2 value of the regression was 96.0%. Validation of this equation resulted in an SEP of 0.063% the regression between the predicted values and the lab values had an R^2 of 82.2% (Figure 1).

The values of the statistics are in line with those found by Smis *et al.* (2014) for their equation for the silica content of *graminoid* species sampled in the alpine parts of the northern, sub-arctic zone of Norway: a calibration equation based on 198 samples and validated with 66 samples resulted in an SEP of 0.102, while the slope of the regression between the predicted values and the lab values was 0.97 and the R^2 was 0.95. By narrowing their data set only to 1 grass species (*Deschampia cespitosa*) the statistics were further improved with a slope of 1.02 and a R^2 of 0.93.

These results suggest that NIRS can be used to predict the silica content both between plant species as within plant species, where the variation in silica is generally lower. Although the equation we developed with the available samples is not suited to determine the silica content very precisely of any tall fescue sample, it showed to be capable to discriminate genotypes with consistently high and low silica contents. For screening populations for breeding purposes this is sufficient. Adding supplementary samples from different trails to the calibration set will make the equation more robust and may eventually lead to a calibration that allows a very precise prediction of the silica content in tall fescue.

Conclusions

NIRS can be used to discriminate between the silica content of different tall fescue genotypes.

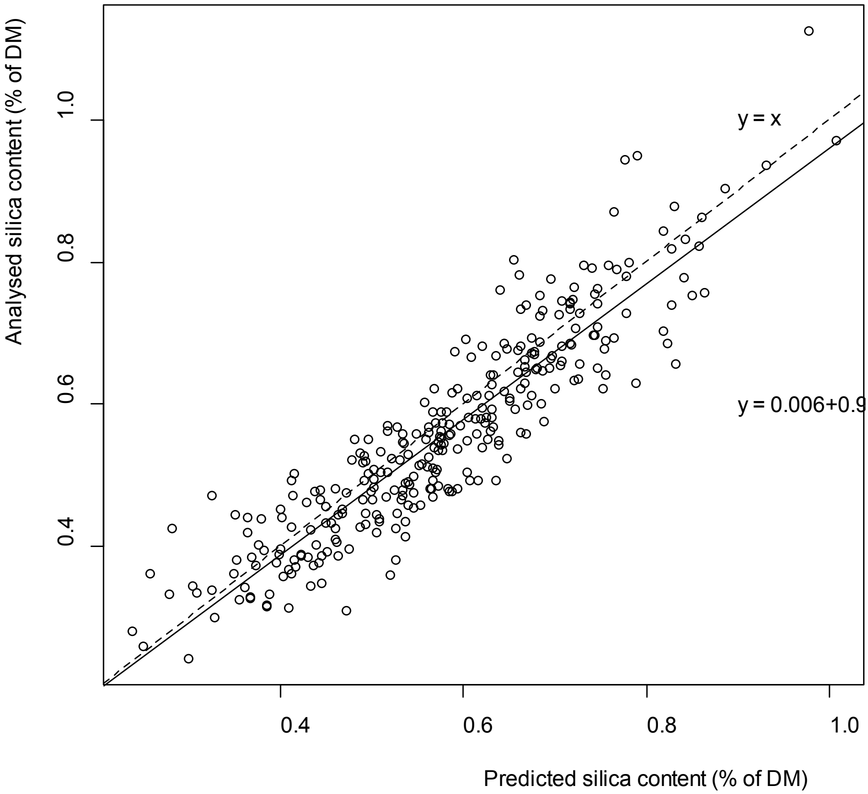

Figure 1. Regression between the silica content of 297 grass samples determined in the lab and predicted using a NIRS calibration equation.

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'WiltExpert' – a model for on-farm prediction of grass wilting time from mowing to ensiling dry matter content

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Abstract

It is recommended to wilt cut grass swards in the field up to a target dry matter (DM) content before ensiling in order to ensure an efficient fermentation and to prevent a poor grass compaction in the clamp. In many German regions, however, less than half of all grass silages meet the recommended DM range. Based on field experiments conducted 1981-1986, a model was developed for on-farm prediction of the wilting time. In order to prepare the on farm use of the model, the objective of the current study was to compare the observed time from mowing till ensiling with the model's prediction under the actual grassland management conditions and to evaluate its predictive ability for northern Germany. To this end, 16 experiments were conducted on 6 farms, where grass samples were taken at mowing and at ensiling to determine their DM content. A Mean Absolute Error (MAE) of 108 minutes (min.) and a modelling efficiency (EF) of 0.99 between observed and predicted time to reach the ensiling DM content reveal that predictive ability of the model was satisfactory.

Keywords: drying process, dry matter content, ensiling, weather conditions

Introduction

Depending on the share of *Lolium* and legume species in a sward, it is recommended to wilt cut grass and legume swards in the field up to a DM content of about 35% to 45% when using choppers and forage wagons and up to 55% when silage is preserved in bales. This ensures an efficient fermentation and prevents a poor grass compaction in the clamp and in the bales. In most northern German regions, however, less than 50% of all grass silages meet the recommended DM content. In particular on larger agricultural farms with a high number of grass fields and an extended silage production period the wilting conditions may vary substantially between single fields. Starting with the DM content at the time of mowing, a reliable prediction of the wilting time up to a target DM content could help the farmers to better meet the DM content requirements on the different fields and to increase silage quality. From 1981 to 1986 harvesting and weather conditions during the field drying processes were studied on more than 1,400 single grass plots at Paulinenaue (52°41' N, 12°43' E, Brandenburg, northeast Germany), representing a wide range of swards. Functional relationships were developed to describe the dynamics of wilting. Based on those field experiments, a model had been developed for on-farm prediction of the wilting time from mowing to ensiling (Rübensam and Bockholdt 1987, Thöns *et al.,* 1989). The developed wilting-model assumes wilting to take place between 9 a.m. and 7 p.m. and describes the time to reach the target DM content by means of empirical regression equations. Initial sward, swath and weather parameters are used as independent parameters (Figure 1). The introduction of the model to farms had started (Thöns and Rübensam 1987) but stopped due to the structural changes in the agricultural advisory system and the farms after the German reunification in 1989. In order to prepare the model use for the on farm prediction of the wilting time, it was the objective of the current study to validate the predictive ability in terms of time required for wilting the grass swaths from mowing till ensiling on farm under actual grassland management conditions of northern Germany.

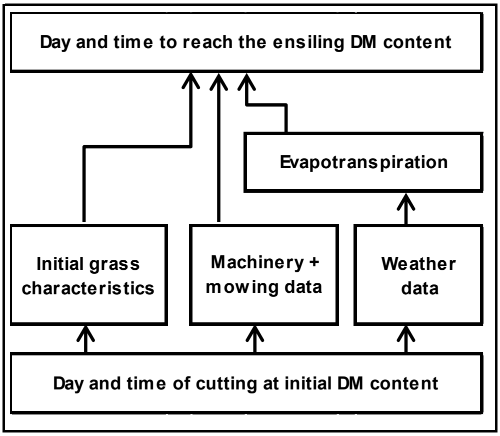

Figure 1. Components of the model 'WiltExpert'.

Materials and methods

The evaluation is based on 16 data sets collected on single fields of 6 farms located in the northern part of Germany (4 Brandenburg, 1 Mecklenburg-Vorpommern and 1 Schleswig-Holstein). Ten data sets originated from the 1^{st} and 6 data sets from the 2^{nd} and 3^{rd} cut. The swards were dominated by *Lolium perenne* and cut at an early stage of growth. The farmers reported the following data to describe the applied harvest process: estimated fresh matter yield, morphological stage of the grasses, type and use of mower, conditioner and tedder, mowing and swath width, mowing and ensiling date and time. Grass samples were taken at the same locations of the field at mowing and at ensiling for DM content determination by oven drying. Weather data (evapotranspiration and precipitation) for the different locations concerned were provided by Germany's National Meteorological Service. For the 16 trials, beside the harvest process and weather data, the observed DM content at mowing, the time of mowing and the observed DM content at ensiling, serving as the target DM content, were used as model inputs. The predicted time required for wilting from observed DM content at mowing till observed DM content at ensiling was compared with the observed wilting time. The predictive ability of the model was evaluated on the basis of the Mean Absolute Error (MAE), the Root Mean Square (RMSE) and the Modelling Efficiency (EF) (Nash and Sutcliffe 1970).

Results and discussion

The period between cutting and ensiling lasted 2,000 minutes (~33 hours) on average. The model was able to satisfactorily predict the wilting time to get an observed DM content of the cut grass swards, as indicated by a MAE of 108 min, an RMSE of 129 min. and an EF of 0.99 (Table 1). A slope of 0.97 not differing from unity and an intercept not significantly different from zero confirms that there is no systematic deviation of predictions (Figure 2). There results were grouped into subsamples, differing with respect to rainfall occurring during the wilting period (with vs without) and with respect to the growing season (first vs second and third cuts). The MAE, RSME and EF values of all different subsamples were within the range of the total sample, again indicating no systematic deviation.

Conclusions

The results reveal that the model was able to predict the time to reach the ensiling DM content with good accuracy. It is suitable for all grassland cuts of the season and rainy periods do not affect prediction accuracy. Based on the results, the model will be developed further and prepared for the practical use by the farmer on-farm. The forecast of evaporation for the different locations will be supplied by Germany's National Meteorological Service and farmers will be instructed to put the different sward and swath

Table 1. Differences between observed and predicted time to reach the DM content at ensiling.[1]

Trials	No.	Rain	DM at mowing	DM at ensiling	Goodness of model fit		
					MAE	RMSE	EF
		mm	g kg^{-1} FM	g kg^{-1} FM	min.	min.	
All	16		220.3	402.1	108	129	0.99
Without rain	8		248.5	448.5	130	147	0.98
With rain	8	3.2	192.1	355.8	87	110	0.99
1st cut	10	2.8	210.2	379.1	103	119	0.99
2nd and 3rd cut	6	6.0	237.2	440.5	118	145	0.99

[1] MAE = mean absolute error; RMSE = root mean square; EF = modelling efficiency.

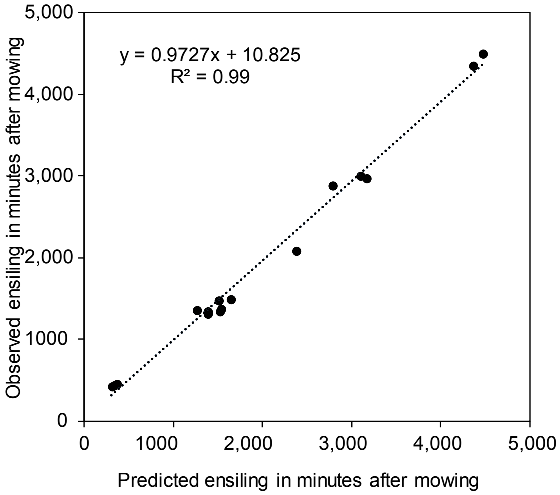

Figure 2. Comparison between observed and predicted time to reach the ensiling DM content.

characteristics of a field correctly into the model. The model could be implemented e.g. as a smart phone app, automatically linked to the weather conditions of the location concerned.

Acknowledgements

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webGRAS: a web application to estimate the potential forage quality of mountain permanent meadows

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Abstract

Knowledge of forage quality is of pivotal importance to farmers in order to provide animals with a suitable ration. The quality of green forage of permanent meadows can be estimated by means of statistical predictive models taking climatic and meteorological variables, as well as additional information about botanical composition, geomorphologic factors, soil properties and management practices into account. Within the ERDF-project webGRAS such predictive models, developed for 19 parameters with a prediction accuracy (R^2) ranging between 0.32 and 0.71, were implemented into a user-friendly web-application, in order to enable farmers and advisors to estimate the forage quality at the first cut of permanent meadows in South Tyrol (Italy). The estimation is partly based on data automatically retrieved from geodatabases and partly on information known to the user, of which the knowledge of the date of the stem elongation stage (15 cm growing height) and the harvest date are the most relevant ones. The application is equipped with a back-end system that generates daily the required meteorological variables from the incoming data of the local weather stations network. A participatory approach, involving relevant local stakeholders and experts, was used along the development of the application to ensure an easy and broad use of the application in practice.

Keywords: forage quality, predictive models, web application, participatory approach

Introduction

Knowledge of forage quality is of pivotal importance to farmers in order to provide animals with a suitable ration. Most reliable and accurate information on forage quality is obtained by means of laboratory analysis, but values can be predicted taking the phenological stage and the botanical composition into consideration (see i.e. Daccord *et al.*, 2007). The quality of green forage of permanent meadows at first cut can be also estimated by means of statistical predictive models taking climatic and meteorological variables, as well as additional information about botanical composition, geomorphologic factors, soil properties and management practices into account (Romano *et al.*, 2016). This quality can be considered to be potential, as it has not been affected by forage conservation processes. The project webGRAS aimed at developing a user-friendly web application implementing such predictive models, enabling farmers and advisors to estimate the forage quality at the first cut of permanent meadows in South Tyrol, Italy.

Materials and methods

The estimation of the quality parameters is based on a database of about 5,700 forage samples, over a time span of 13 years in 202 environments (site × year combinations) and describing changes over time in the forage quality of permanent meadows at the first cut for seven weeks starting from the stage of stem elongation. In this way, the starting point for the description of the course of forage quality is bound to an attribute, which is easily identifiable by practitioners. Statistical predictive models for 19 quality parameters have been developed by means of mixed models according to Romano *et al.* (2016). They make use of meteorological (growing degree-days, daily precipitation departures from the long term average within homogenous precipitation districts) or climatic (sum of topographically computed, potential solar radiation) variables. Variables related to geomorphology, botanical composition, agronomic management and soil (results of a soil analysis) are taken into account as well. In order to ensure the functioning of the application even with partially missing information, four models taking different sets of independent variables into account were developed for each quality parameter: (1) all variables; (2) only variables, the value of which is presumably known to most of the users; (3) all variables but the results of a soil analysis; (4) all variables but the meadow type. The application is equipped with a back-end system that generates daily the required meteorological variables from the incoming data of the weather stations network of the Autonomous Province of Bolzano. The relevant local stakeholders and experts on forage production and animal feeding have been systematically involved during the development of the application in order to account for their expectations, knowledge (i.e. the appraisal of the information known to most of the users) and for their inputs concerning several aspects of the application (i.e. workflow, layout of input masks, contextual helps) and ensure an easy and broad use of the application in the practice.

Results and discussion

Predictive models have been developed for a total of 19 forage quality parameters with a prediction accuracy (R^2) ranging between 0.33 and 0.7 (Figure 1). In general, ash and minerals are less accurately predicted than the other parameters, but the information provided by soil analyses and meadow type contributes to improve the accuracy. For all other parameters accuracy is already satisfactory using only the obligatory variables.

These models have been integrated into a user-friendly software freely accessible on the web (webgras.laimburg.it). The estimation is partly based on data automatically retrieved from geodatabases and partly on information known to the user, of which the knowledge of the date of the stem elongation stage (15 cm growing height) and the harvest date are the most relevant ones. The operative workflow is described in Figure 2. At step 1 the user identifies the meadow on a GIS-interface either by a cadastral query or with simple drawing tools on the background of ortophotos and cadastral maps. From this information the respective precipitation district is assigned and the mean topographic features are computed (step 2). At step 3, information is required from the user. He is allowed to provide two notional harvest dates along with the real one. For the botany-related variables, contextual, interactive, user-friendly helps are integrated into the application. The date of stem elongation and the harvest date are used in combination with the daily potential global radiation (step 4), with the daily precipitation departures from the long term average within the respective district (step 5) and with the interpolated mean daily temperatures

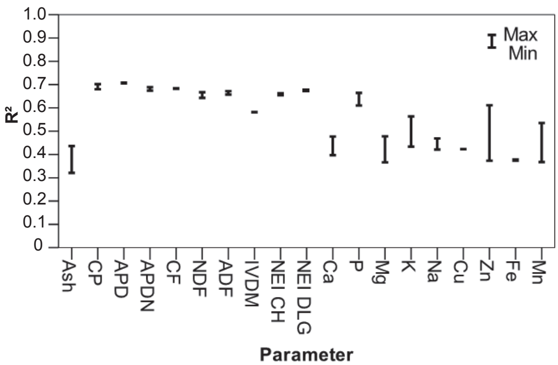

Figure 1. Summary of the prediction accuracy of the statistical models (squared correlation between predicted and observed value of a five-fold cross-validation). For each parameter the highest value refers to the model taking all variables into account. CP = crude protein; APD = absorbable intestinal protein; APDN = absorbable intestinal protein based on the nitrogen available in the rumen; CF = crude fiber; NDF = neutral detergentfiber; ADF = acid detergent fiber; IVDM = in vitrodigestibility; NEl CH = net energy for lactation (Swiss system); NEl DLG = net energy for lactation (DLG-system).

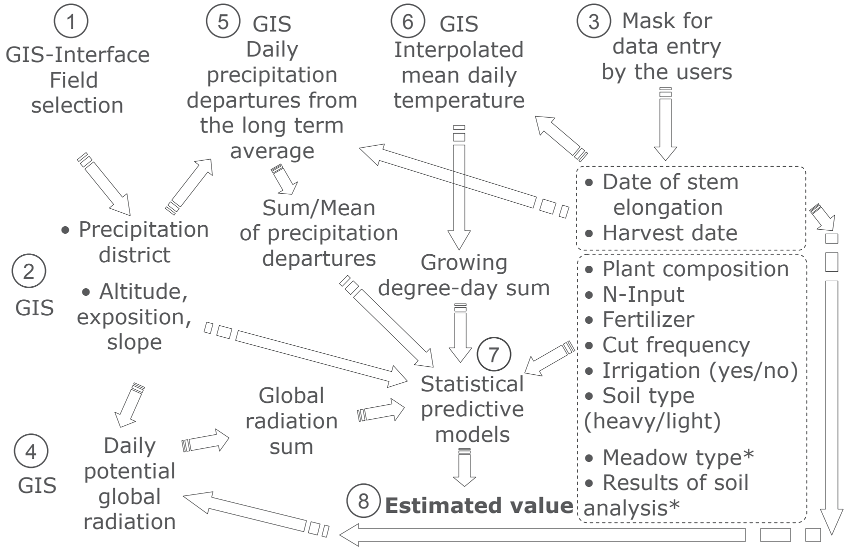

Figure 2. Workflow of the web application. * indicates facultative data entry by the users.

(step 6) to compute the necessary meteorological and climatic variables. Together with all other variables they are used to feed the predictive statistical models (step 7), which provide estimated values for the different parameters of the forage quality (step 8). The user obtains a report summarising the predicted forage quality for both the real and the notional harvest dates, in order to increase the user's awareness for change in the forage quality depending on the harvest timing. Moreover, the system allows correcting the input data of existing reports and using existing reports as a basis for new reports, minimising the time effort for new report requests.

Conclusions

Since spring 2016, the web application has been available to advisors and farmers for estimating the forage quality at the first cut of permanent meadows in South Tyrol.

Acknowledgements

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The in-situ ruminal degradation characteristics of a perennial ryegrass and mixed perennial ryegrass and white clover swards

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Abstract

White clover (*Trifolium repens* L.; clover) can play an integral part in pasture based animal production systems due to its increased crude protein (CP) and reduced structural fibre concentrations. The objective of this experiment was to measure the *in-situ* ruminal degradation of CP and neutral detergent fibre (NDF) of a perennial ryegrass (*Lolium perenne* L.; PRG)-clover sward receiving 150 kg N ha^{-1} yr^{-1} (Cl150), a PRG-clover sward receiving 250 kg N ha^{-1} yr^{-1} (Cl250), and a PRG sward receiving 250 kg N ha^{-1} yr^{-1} (Gr250). Samples were collected prior to grazing during the 2013 grazing season, freeze-dried and milled through a 1 mm screen. Samples were bulked by rotations 1 to 5 (February – June; T1) and rotations 6 to 9 (June – November; T2) for each treatment. In T1 the Cl150 treatment had a lower (P<0.01) Dg_{CP} than Cl250 and Gr250, but in T2 the Gr250 treatment had a lower (P<0.01) Dg_{CP} than Cl150 and Cl250. Cl150 had a greater (P<0.05) Dg_{NDF} than Gr250, and the lowest NDF concentration of all three treatments. In conclusion, including clover in a PRG sward increased Dg_{CP} in T2 when sward clover proportion was >290 g kg^{-1} DM, and increased Dg_{NDF} compared to a PRG only sward.

Keywords: perennial ryegrass, white clover, rumen degradability, CP, NDF

Introduction

Perennial ryegrass (*Lolium perenne* L; PRG) is considered to be the most important grass species in temperate climates due to its high digestibility and good grazing tolerance (Wilkins, 1991). White clover (*Trifolium repens* L.; clover) can play an integral part in pasture based animal production systems, due to its greater nutritive value compared to other sown sward species (Ulyatt *et al.,* 1977) which is related to its high crude protein (CP) concentration (Van Soest, 1994) and lower total cell wall or structural fibre concentration compared to PRG (Thomson, 1984). The inclusion of clover into a PRG sward can increase herbage CP concentration (Van Soest, 1994) and reduce fibre concentration (Thomson, 1984) which can positively influence voluntary dry matter (DM) intake and animal performance (Thomson, 1984). The largest increase in animal performance on PRG-clover swards usually occurs in the second half of the growing season (Egan *et al.*, 2014). The objective of this experiment was to measure the *in situ* degradation of CP and neutral detergent fibre (NDF) of a grazed PRG-clover sward receiving 150 kg N ha^{-1} yr^{-1} (Cl150), a grazed PRG-clover sward receiving 250 kg N ha^{-1} yr^{-1} (Cl250) and a PRG only sward receiving 250 kg N ha^{-1} yr^{-1} (Gr250).

Materials and methods

Paddocks were grazed 9 times between February and November 2013 to a post-grazing sward height of 4 cm. Herbage samples were collected from each paddock to a stubble height of 4 cm immediately prior to grazing. Swards had on average a regrowth interval of 24 days at sampling, with a pre-grazing herbage mass of 1,448±90.5 kg DM ha^{-1}. Samples were freeze-dried at -55 °C for 72 h, then milled through a 1 mm screen, and then bulked by sward type and rotation, from rotations 1 to 5 (time stage 1 (T1) – February to early June) and rotations 6 to 9 (time stage 2 (T2) – June to November). Clover proportion of Cl150 and Cl250 was determined once per paddock in each rotation before grazing as described by Egan *et al.* (2014). Three mid-lactation Holstein Friesian dairy cows permanently fitted with rumen cannulae were used to incubate the samples in January 2014. Animals were housed and offered a 72:28 forage:concentrate diet. Two g of herbage sample was placed in nylon bags and heat sealed. All bags were incubated in the rumen at 08:30 h for 0, 3, 8, 12, 24, 72 and 240 h. After removal, nylon bags were washed through two 15 min cold wash cycles, then oven dried at 60 °C for 48 h, and then weighed and analysed for CP and NDF. The immediately soluble fraction (A) for CP (A_{CP}) and NDF (A_{NDF}) was measured as the fraction released after washing the 0 h bags. The residue remaining after 240 h of incubation was assumed to be the undegradable fraction (C) of CP (C_{CP}) and NDF (C_{NDF}). The degradation curve of the insoluble but potentially degraded fraction B (C – A) was fitted by a first order model described by (Ørskov and McDonald, 1979), including A, C, B and the rate per h of degradation of B (Kd). Rumen degradability (Dg) of CP (Dg_{CP}) and NDF (Dg_{NDF}) in the rumen was calculated assuming a fractional passage rate (*kp*) of 0.060 h^{-1}. Rumen degradability of CP and NDF were analysed using the PROC MIXED procedure in SAS (SAS Institute, 2003). Sward type, time stage, cow and the associated interaction between sward type and time stage were included in the model.

Results and discussion

Clover proportion of Cl150 and Cl250 was similar in T1 (146 g kg^{-1} DM ± 9.2). In T2 Cl150 had a greater clover proportion than Cl250 (389 and 295 g kg^{-1} DM ± 13.7, respectively). Chemical composition of the samples placed in the rumen is shown in Table 1. There was an effect of the sward type × time interaction ($P<0.01$) on Dg_{CP} (Table 2). Cl250 and Gr250 had the greatest CP concentration in T1 and the greatest Dg_{CP}. In T2 Cl150 and Cl250 had the greatest CP concentration and greatest Dg_{CP}. The greater Dg_{CP} on Cl250 and Gr250 (+55 kg N ha^{-1}) compared to Cl150 in T1 was due to the higher N fertiliser application (Salaün *et al.,* 1999). Nitrogen fertiliser increases the CP concentration of the sward. Harris *et al.* (1997) showed that sward clover proportion has a similar effect on sward CP

Table 1. Crude protein and neutral detergent fibre concentration of a PRG-clover sward receiving 150 kg N ha^{-1} yr^{-1} (Cl150), a PRG-clover sward receiving 250 kg N ha^{-1} yr^{-1} (Cl250) and a PRG only sward receiving 250 kg N ha^{-1} yr^{-1} (Gr250) during February to June (Time stage 1) and during June to November (Time stage 2).

	Time stage 1			Time stage 2		
	Cl150	Cl250	Gr250	Cl150	Cl250	Gr250
Crude protein (g kg^{-1} DM)	175	205	191	214	183	175
Neutral detergent fibre (g kg^{-1} DM)	287	324	277	301	301	344

Table 2. Forage crude protein (CP) degradation characteristics (% of DM)[1] as defined by soluble fraction (A), potentially degradable fraction (B), undegradable residue (C), fractional rate of degradation (Kd h-1), and effective degradability (Dg), of a PRG-clover sward receiving 150 kg N ha^{-1} yr^{-1} (Cl150), a PRG-clover sward receiving 250 kg N ha^{-1} yr^{-1} (Cl250) and a PRG only sward receiving 250 kg N ha^{-1} yr^{-1} (Gr250) during February to June (Time stage 1) and during June to November (Time stage 2).

	Time stage 1			Time stage 2				Level of significance		
Item	Cl150	Cl250	Gr250	Cl150	Cl250	Gr250	SED[2]	Sward type	Time Stage	Sward type × Time Stage
A_{CP}	0.677^{a}	0.845^{c}	0.787^{bc}	0.748^{ab}	0.799^{bc}	0.781^{bc}	0.0239	0.01	0.75	0.09
B_{CP}	0.303^{a}	0.148^{b}	0.204^{bc}	0.231^{c}	0.182^{bc}	0.206^{bc}	0.0218	0.01	0.52	0.09
C_{CP}	0.020	0.007	0.010	0.022	0.019	0.014	0.0026	0.01	0.05	0.15
Kd_{CP}	0.133	0.106	0.124	0.150	0.143	0.060	0.0301	0.2	1.00	0.24
Dg_{CP}	0.886^{a}	0.939^{b}	0.923^{bc}	0.911^{c}	0.917^{c}	0.885^{a}	0.0068	0.01	0.06	0.01

[1] Measured from 0 to 240 hours.

[2] SED = standard error of the difference between means.

concentration which can be seen in T2 in the current study where CP concentration was greater in Cl150 and Cl250 than Gr250 (199 and 175 g kg^{-1} DM, respectively), despite Cl150 receiving 45 kg N ha^{-1} less than Cl250 in T2. Salaün *et al.* (1999) found that an increase in the use of N fertiliser increased the solubility of CP in forages. This was reflected in the current study where there was a tendency for an effect of the sward type × time interaction (P=0.09) on the soluble A_{CP} fraction. In T1 the greater application of N fertiliser increased the soluble A_{CP} fraction on Cl250 and Gr250 compared to Cl150 which led to an increase in the B_{CP} on Cl150 in T1. Sward type and time stage had a significant effect on the insoluble C_{CP} fraction (P<0.05). Salaün *et al.* (1999) found that the C_{CP} was influenced by level of N fertiliser applied. As the N fertiliser was reduced in Cl150, C_{CP} increased. Sward type did not have a significant effect on Kd_{CP} (P>0.05). The data presented for NDF (Table 3) are for rumen incubation times 0, 3, 8 and 12 h, as there was insufficient sample for chemical analysis remaining after incubation times 24, 72 and 240 h. Sward type had a significant effect (P<0.05) on Dg_{NDF}, with Cl150 greater than Gr250 and Cl250 intermediate. There was a tendency (P=0.08) for sward type to have an effect on B_{NDF}. The Cl150 had the greatest and Cl250 had the lowest potential degradable B_{NDF} fraction. This could be accounted for by the low A_{NDF} of Cl150 and high A_{NDF} of Cl250 and similar C_{NDF} fraction among the three sward types.

Table 3. Forage neutral detergent fibre (NDF) degradation characteristics (% of DM)[1] as defined by soluble fraction (A), potentially degradable fraction (B), undegradable residue (C), fractional rate of degradation (Kd h^{-1}), and effective degradability (Dg) of a PRG-clover sward receiving 150 kg N ha^{-1} yr^{-1} (Cl150), a PRG-clover sward receiving 250 kg N ha^{-1} yr^{-1} (Cl250) and a PRG only sward receiving 250 kg N ha^{-1} yr^{-1} (Gr250) during February to June (Time stage 1) and June to November (Time stage 2).

	Sward type					Time stage			
Item	Cl150	Cl250	Gr250	SED[2]	P-value	Time stage 1	Time stage 2	SED	P-value
A_{NDF}	0.265	0.325	0.261	0.0332	0.36	0.308	0.260	0.0271	0.23
B_{NDF}	0.421	0.292	0.342	0.0364	0.08	0.327	0.376	0.0297	0.27
C_{NDF}	0.314	0.384	0.397	0.0261	0.14	0.365	0.365	0.0213	1.00
Kd_{NDF}	0.169	0.176	0.268	0.0593	0.38	0.218	0.190	0.0484	0.67
Dg_{NDF}	0.574[a]	0.543[ab]	0.514[b]	0.0201	0.05	0.545	0.542	0.0164	0.23

[1] Measured from 0 to 12 hours.

[2] SED = standard error of the difference between means.

Conclusions

Sward type had an effect Dg_{CP}, which can be attributed to an increase in CP concentration. In T1 Cl250 and Gr250 had the greatest Dg_{CP} as a result of the higher level of N fertiliser applied and increased CP concentration compared to Cl150. However when sward clover content increased in T2 (>290 g kg^{-1} DM), Dg_{CP} was greater on Cl150 and Cl250 compared to Gr250. The NDF degradability was also affected by sward type. The Cl150 had the greatest Dg_{NDF} and Gr250 the lowest, measured up to 12 h incubation in the rumen. The Dg_{NDF} should be quantified for the full 240 h period.

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Prediction of the in vivo organic matter digestibility of cereal-legume intercrops silages

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Abstract

There is an increasing interest in using cereal-legume intercrops mixtures in ruminant production systems to assist forage self-sufficiency and climate change adaptation. Available data on the energy value of the cereal-legume silages are limited due to a lack of *in vivo* organic matter digestibility (OMD) measurements and the potential vast number of mixtures used in practice. The objective of this work was to study the relationships between *in vivo* OMD and some chemical and enzymatic parameters of different cereal-legume silages in order to identify a potential predictor for the OMD. Data from four trials with sheep carried out on the experimental farms of INRA Le Pin-au-Haras (France) and Agroscope, Posieux (Switzerland) were used. The cereals in the mixtures were oat, wheat or triticale, and the legumes were pea and vetch. Mixtures were harvested at two stages of growth. Significant relationships were observed between OMD and crude protein content, legume proportion and pepsine-cellulase digestibility (PCD). The best model to predict OMD used PCD (R^2=0.71). Additional data with other mixtures and legume proportions are required to confirm this result, but PCD would enable the prediction of *in vivo* OMD of cereal-legume silages with good precision.

Keywords: *in vivo* digestibility, cereal-legume silages, prediction

Introduction

Intercropping cereals with legumes is developing in French ruminant production systems to permit forage self-sufficiency, reduce nitrogen fertilizers and for climate change adaptation. Silages produced from a mixture of whole-crop cereal and whole-crop legume are likely to have a higher concentration of crude protein (CP) and digestibility than cereal monoculture or moderate quality grass silage (Adesogan *et al.*, 2002; Adesogan *et al.*, 2004; Pursiainen and Tori, 2006). However, the feed value of these silages is still not well-known due to a lack of *in vivo* measurements and the potential vast number of mixtures used. Few references are available in the Feed Tables from European countries. In France, no predictive equation is available to estimate the *in vivo* organic matter digestibility (OMD) for these silages from common predictors such as *in vitro* digestibility or the chemical composition of silages. The *in vivo* OMD of the cereal-legume silages is, therefore, estimated using predictive equations developed for other forages such as grass, legume or maize silages (INRA, 2007), which is certainly not adapted to these forages. The objective of this work was to study the statistical relationships between *in vivo* OMD and a few chemical and enzymatic characteristics of cereal-legume silages in order to find a method to estimate OMD suitable for these forages.

Materials and methods

Data (n=16) from *in vivo* experimentations carried out between 2010 and 2014 in two experimental farms: Le-Pin-au-Haras, INRA, France (two digestion trials) and Posieux, Agroscope, Switzerland (two digestion trials), were used. In the INRA first trial, the silages tested were field pea (*Pisum sativum* L.) + common vetch (*Vicia sativa* L.) with the cereal wheat (*Triticum aestivum* L.), beardless triticale (*X Triticosecale)* or bearded triticale at the seed ratio of respectively 17, 20 and 220 seeds m^{-2}. These three silages were harvested at two stages of growth: early (about 20% dry matter, DM, at harvest) and late (about 30% DM at harvest). In the INRA second trial, the silages were field pea + wheat harvested at the two same stages of growth. Silages tested at Agroscope were field pea + triticale + oat (*Avena Sativa* L.) harvested at late stage of growth (cereals at dough-milky stage); silages differed in the pea to cereals seed ratio (Arrigo, 2014; Arrigo *et al.,* 2015). The different silages were ensiled without additives except for three silages that received Kofasil Bale® (Addocon, Germany) and after wilting for mixtures harvested at an early stage.

In all experiments, *in vivo* OMD for each silage was measured on 4 to 6 male castrated sheep. The botanical composition of the forages was determined at harvest. Silages were analysed for ash, CP content (Dumas method, Rapid N Cube, Elementar Analysensystem GmbH), neutral detergent fibre (NDF) and acid detergent fibre (ADF) contents using an Ankom fiber analyser (Ankom Technology, Macedon, NY, USA) with heat-stable amylase. Pepsin-cellulase digestibility (PCD expressed as g kg^{-1} DM) of the forages was determined according to Aufrère and Michalet-Doreau (1983). The statistical relationships (linear and stepwise regressions) between OMD and the chemical composition, legume proportion and PCD of the silages were tested with the Minitab® statistical software.

Results and discussion

Descriptive statistics on chemical composition and digestibility of the silages are shown in Table 1. The DM content was on average 309 g kg^{-1}, the silages from Switzerland being dryer (351 g kg^{-1} DM) than silages from France (273 g kg^{-1} DM). The CP content varied from 54 to 161 g kg^{-1} DM and ADF content from 302 to 398 g kg^{-1} DM. On average, the *in vivo* OMD was 638±40.3 g kg^{-1} OM, which was consistent with previous *in vivo* OMD measurements on cereal-legume silages (Adesogan *et al.*, 2002). The legume proportion at harvest was low (0.17 on average), only 5 mixtures had a legume proportion higher than 0.20.

The CP content (P<0.001, R^2=0.70) and OMD (P=0.02, R^2=0.27) of silages increased with the increase in the legume proportion in the mixture, whereas the NDF content decreased. The ADF content did not differ with the legume proportion in the mixture. On average, the CP content and OMD were slightly higher for the silages harvested at early stage (108 g kg^{-1} DM and 666 g kg^{-1} OM) than for the silages harvested at late stage (91 g kg^{-1} DM and 628 g kg^{-1} OM).

Significant relationships were obtained between *in vivo* OMD and the legume proportion in the mixture, CP content (Equation 1) and PCD (Equation 2). The type of mixture and the *in vivo* data origin (France vs Switzerland) were not significant.

$$\text{OMD (g kg}^{-1}\text{ OM)} = 1.039^{(P=0.001)} \times \text{CP (g kg}^{-1}\text{ DM)} + 539^{(P<0.001)} \quad (1)$$

with n_{data} = 16, RMSE = 26.7 and R^2 = 0.56.

Table 1. Chemical composition and digestibility of the cereal-legume silages (n = 16) used to establish a predictive equation of the *in vivo* organic matter digestibility.

	Mean	S.D.	Min	Max
Legume proportion	0.17	0.24	0	1
Dry matter (DM), g kg^{-1}	309	62.1	207	423
Crude protein, g kg^{-1} DM	95	29.8	54	161
Neutral detergent fibre, g kg^{-1} DM	533	46.2	441	649
Acid detergent fibre, g kg^{-1} DM	329	23.5	302	398
Ash, g kg^{-1} DM	66	16.8	47	104
Pepsine-cellulase digestibility, g kg^{-1} DM	545	62.7	421	662
In vivo organic matter digestibility, g kg^{-1} OM	638	40.3	583	715

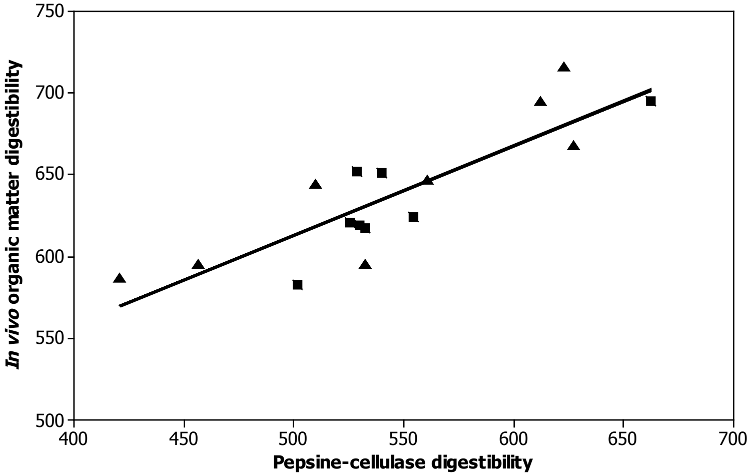

Figure 1. The relationship between *in vivo* organic matter digestibility (g kg^{-1} OM) and pepsine-cellulase digestibility (g kg^{-1} DM) of cereal-legume silages (▲: data from INRA, France; ■: data from Agroscope, Switzerland).

$$\text{OMD (g kg}^{-1}\text{ OM)} = 0.548^{(P<0.001)} \times \text{PCD (g kg}^{-1}\text{ DM)} + 339^{(P<0.001)} \quad (2)$$

with n_{data} = 16, RMSE = 21.6 and R^2=0.71.

The best model to estimate OMD used PCD as the predictive variable (Figure 1). Inclusion of CP content or legume proportion in Equation 2 did not improve the prediction.

The RMSE of the Equation 2 was low (21.6 g kg^{-1}). However, the R^2 is slightly lower than R^2 of equations previously developed for others forages (0.71 vs 0.83 for grass or legume silages and 0.78 for grass; INRA, 2007). These differences could be explained by the limited number of data in this study, or by a different pattern of seasonal OMD changes for cereal-legume crops compared with grass or legume crops.

Conclusions

The equation using the *in vitro* PCD as predictor allows an estimation of the OMD of cereal-legume silages samples with a good precision. However, additional data with others cereal and legume species and different legume to cereal proportions, especially silages performed from crops with greater proportion of legume (>0.35) are required to confirm these results.

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Comparison of protein degradation in the rumen measured in situ and in vivo

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Abstract

The aim of this study was to compare whether the effect of increased dry matter (DM) concentration in grass-clover silage on rumen protein degradation measured *in situ* reflect the actual *in vivo* change in protein degradation in the rumen. Eight grass-clover silages with DM concentrations ranging from 283 to 725 g kg^{-1} were fed *ad libitum* as the sole feed to four rumen and duodenal fistulated Holstein dairy cows in a crossover design. Based on duodenal samples, three external markers and purine concentration in isolated rumen microbes, microbial protein flow at the duodenum and true rumen digestibility of feed protein were estimated. Additionally, the silages were incubated in the rumen (*in situ*) of three cows in Dacron bags with eight incubation times, and the effective protein degradation was calculated assuming a rate of passage of 0.08 h^{-1}. The protein degradation was reduced with 36 and 44 g kg^{-1} when increasing the silage DM concentration with 100 g kg^{-1} for *in situ* and *in vivo* measurements, respectively. The slope from regression of *in vivo* on *in situ* was 0.91, but not significantly different from one. In conclusion, *in situ* measurements reflect the *in vivo* change in true rumen protein degradation well.

Keywords: protein degradation, *in situ*, *in vivo*, forage

Introduction

The amount of absorbed protein is a major factor affecting the milk production in dairy cows (Allen, 1996). Therefore it is important to have a high and stable forage production with a high protein quality. Most forage used for milk production is conserved as silage, but the dry matter (DM) concentration in the plant material before ensiling will affect the microbial fermentation during the ensiling process. A higher DM concentration reduces the microbial fermentation, by which the final silage has a higher concentration of sugar and true protein and a lower concentration of fermentation products (Harrison *et al.*, 1994). When feeding this silage, the microbial synthesis in the rumen will increase and a higher proportion of undegraded feed protein will reach the small intestine (Johansen and Weisbjerg, 2015). In feed evaluation systems most parameters are determined by *in situ* or *in vitro* techniques and it is important that these techniques reflect what actually happen *in vivo* when they are used to optimise feed rations. Despite widespread use of the *in situ* technique very few *in situ*-*in vivo* evaluations have been published. Therefore, the objective of this study was to test whether the reduced rumen protein degradation with increased DM concentration in grass-clover silage measured *in situ* reflect the actual *in vivo* change in protein degradation in the rumen.

Materials and methods

Spring growth and first regrowth of grass-clover swards grown by two Danish organic farmers were cut and pre-wilted to a planned DM concentration of 350 and 700 g kg^{-1}, resulting in total eight silages ensiled without additives and with DM concentrations ranging from 283 to 725 g kg^{-1}. For *in vivo* determination of rumen degradation of crude protein (CP) two primiparous and two multiparous Holstein dairy cows (551±33 kg body weight, 216±23 days in milk, mean ± SD) fistulated in rumen and duodenum were fed *ad libitum* with the silages in a crossover design, with 5 periods of 3 weeks. No concentrate was offered, but minerals and vitamins were offered daily. Three markers (10 g Cr_2O_3, 10 g TiO_2 and 2 g $YbCl_3{\cdot}6H_2O$) were dosed in the rumen twice a day for measurement of flow at duodenum.

In the last week of each period daily feed intake was registered and 12 subsamples from the duodenum were collected over 94 h to cover the diurnal variation, pooled, and subsequently analysed. Once in each period, microbes were isolated from the rumen fluid and analysed for CP and purines to estimate the flow of microbial protein at the duodenum. The flow of nutrients in the duodenum was calculated based on the average of the concentration of each marker in relation to daily supply. True rumen digestibility of CP was calculated as feed intake minus duodenal flow corrected for microbial contribution divided by feed intake. It was assumed that the endogenous flow of CP in the duodenum was constant between treatments and therefore not corrected for. Statistical analyses were done in R (R Core Team, 2014) using a linear random regression model with DM concentration as fixed effect, cow and growth × farmer as random intercepts and with a growth × farmer random slope.

Additionally, the silages were dried, milled (1.5 mm cutter mill) and weighed out in Dacron bags (pore size: 38×38 µm, effective size: 10×7.5 cm) for *in situ* determination of rumen degradation of CP, and incubated in the rumen for 0, 2, 4, 8, 16, 24, 48 and 96 hours, respectively. For each of the silages, one bag was used for each incubation time and repeated in three fistulated dry cows fed at maintenance. After incubation all samples were washed, dried and analysed for the concentration of CP. Using non-linear regression in R (R Core Team, 2014) degradation profile parameters (Ørskov and McDonald, 1979) for each silage were estimated according to the following equation:

Degradation profile (t) = $a + b(1 - e^{ct})$,

where a is the soluble fraction, b is the insoluble but rumen degradable fraction, c is the fractional rate of degradation of fraction b (h^{-1}) and t is incubation time (h). Based on the estimated parameters the effective protein degradation (EPD) was calculated for each of the silages by the following equation (Ørskov and McDonald, 1979):

$EPD = a + b(c / c+k)$,

where k is the fractional rate of passage out of the rumen set to 0.08 h^{-1}, as no correction for particle loss was made. A linear regression with DM concentration as fixed effect was performed.

The *in vivo* degradability were compared with the calculated EPD and the estimated parameters a, b and c in a linear random regression model with EPD, a, b or c as fixed effect, cow and growth × farmer as random intercepts and with a growth × farmer random slope.

Results and discussion

The DM intake of the cows (12.5±2.1 kg d^{-1}, mean ± SD) was not affected by the DM concentration in the silage (P=0.6) in the *in vivo* experiment. It is therefore assumed that the rate of passage out of the rumen was not affected either. The true rumen degradation measured *in vivo* decreased with 44 g kg^{-1} (P=0.02) when increasing the DM concentration in the silage with 100 g kg^{-1} (Figure 1). In comparison, the EPD measured *in situ* decreased by 36 g kg^{-1} (P<0.001) when increasing the DM concentration in the silage with 100 g kg^{-1}. The measured EPD values varied from 715 g kg^{-1} in the silage with the highest DM concentration to 884 g kg^{-1} in the silage with lowest DM concentration. As seen in Figure 1, the actual values for the true rumen digestibility measured *in vivo* were considerable smaller than the EPD values. This indicates that the contribution of CP from endogenous sources was high in the duodenal flow, which probably partly was due to the relatively low feed intake.

The regression of the true protein degradation measured *in vivo* on EPD measured *in situ* was significant (α=0.91, P=0.02) and did not differ from one (P=0.8) showing that the change in protein degradation

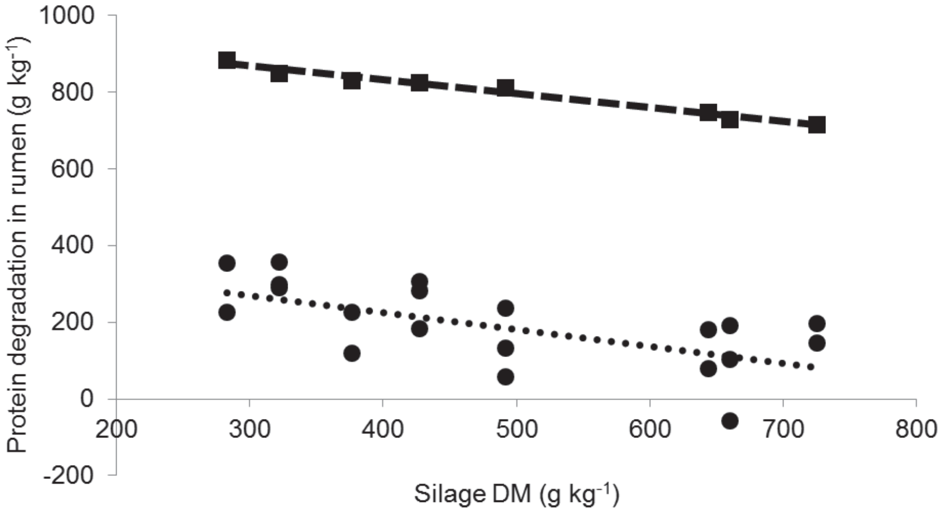

Figure 1. True rumen protein degradation determined *in vivo* (●, n=20) with the regression line (••••, y = −0.44 + 401) and the effective protein degradation determined *in situ* (■) with the regression line (▬ ▬, y = −0.36x + 976).

measured *in vivo* is reflected well in *in situ* measurements. The *in vivo* degradability were positively correlated to the soluble fraction a (P=0.02), negatively correlated to the insoluble but degradable fraction b (P=0.03) but not correlated to the fractional rate of degradation (P=0.6). When correcting the degradation parameters for particle loss, estimated as the difference between zero hour solubility and solubility over filter paper, and using 0.05 h^{-1} passage rate for calculation, the regression coefficient between *in vivo* and *in situ* measurements was close to one, but not significant (α=0.97, P=0.17).

Conclusions

It is concluded that the change in rumen protein degradation, when increasing the DM concentration of grass-clover silage measured *in vivo,* is well reflected in the EPD measured *in situ*.

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Assessments of the values of multi-species grassland for grazing, silage and hay production

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Abstract

Different grassland utilizations are possible: direct utilization by animals (grazing) or forage harvest and conservation (either by ensilage or hay). The grasslands value for these different utilizations depends on many variables of the vegetation. These variables can be aggregated to propose scores for the grazing, ensilage and hay. We developed and used these indicators on four annual mixtures from the seed company FERTIPRADO® set up in south of France. Each of these mixtures is composed of 6 legumes and 2 grasses and managed with no irrigation and no nitrogen fertilizer. Botanical composition, height, and nutritional contents, measured using near infrared spectrometry (NIRS) directly on the sward, were monitored every two weeks. From these variables, indicators were developed using the aggregation tool: TATALE. With TATALE, the input variables are first transformed into scores between 0 and 1 and secondly these different scores are aggregated to obtain summary scores. The parameterization (transformation and aggregation parameters) was made by expertise and literature surveys.

These tools provide scores for grazing, silage and hay production values of grassland based on the vegetation state. To illustrate these results, we compared the evolution of one of the four mixtures during the season.

Keywords: multi-specific sward, forage quality, assessment tools

Introduction

Agronomical value of grasslands relies on the quantity of biomass produced and the qualities of the forage. The quality of forage is generally described by several criteria: The protein content and energy concentration and the digestibility. The quantity and the quality of the forage change during the season. Furthermore, the assessment of the agronomical value may also depend of the way the grassland is used. Indeed, farmers can use grassland to feed their animals in different ways: The animals can directly graze the forage or the grass is mown by the farmers. The mown grass can be stored according to two processes: silage or hay. For a given sward, its values for the different uses could be different. A tall sward is less suitable for grazing (animals are not able to eat the tall plants). However a tall sward is interesting for mowing and hay (more standing biomass).

In this paper, we will propose a method to develop indicators to evaluate the value of grasslands for the different uses: grazing, ensilage and hay. We develop and test these indicators on the legume rich mixtures proposed by FERTIPRADO Corporation. The work presented here is a work in progress.

Materials and methods

In the Melgueil experimental site (close to Montpellier, south of France), we tested in 2013, four mixtures proposed by the seed company FERTIPRADO®. These mixtures of 8 annual species were rich in legumes (6 species). The grasslands were managed with only phosphorus and potassium application and with one cut during the year. Every second week, the botanical composition and the height of the grass were assessed. NIRS were used on the standing vegetation to assess the chemical composition (digestibility, fibre and nitrogen content). At the end of the season, the yield was measured. We assumed that the

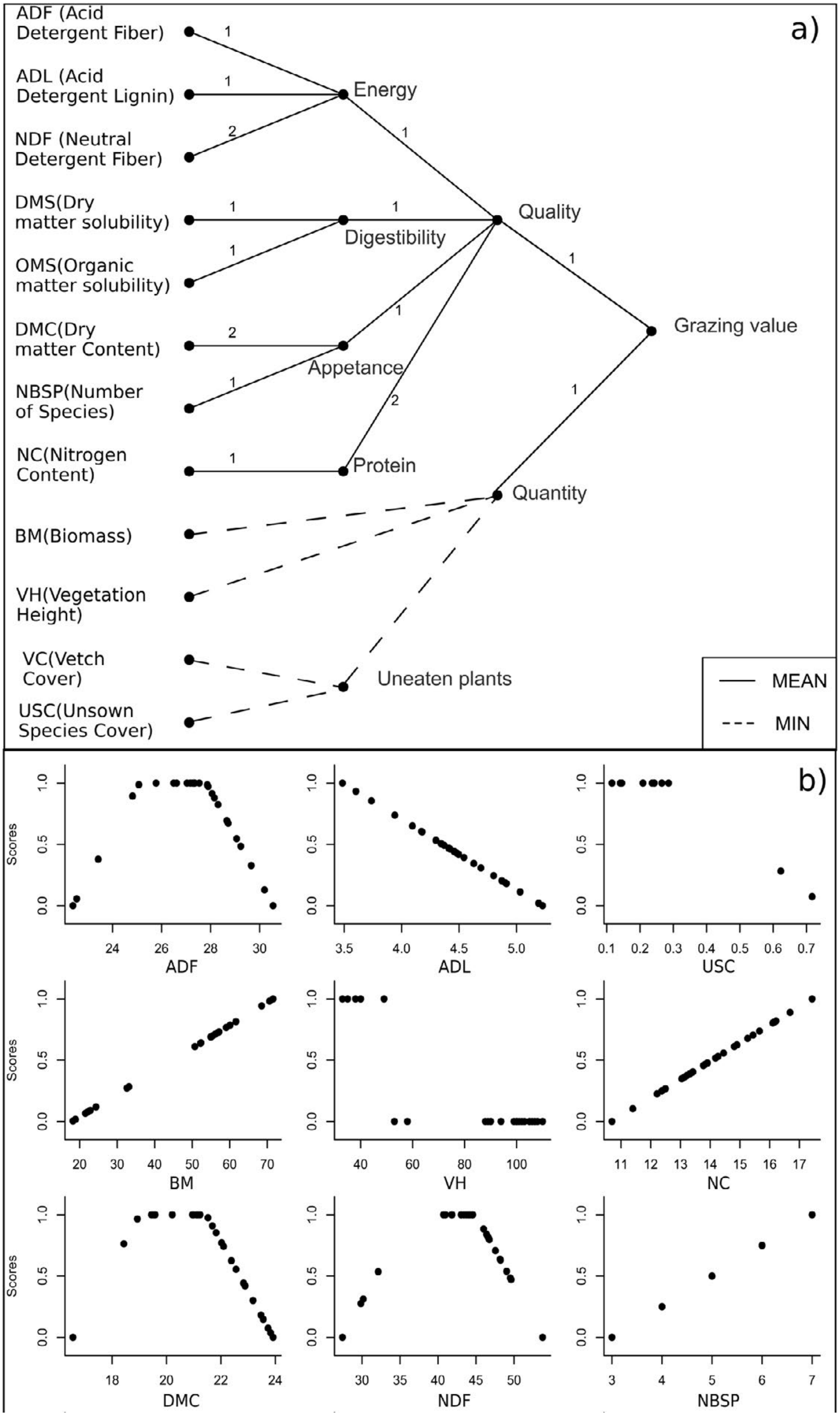

Figure 1. (A) Aggregation tree for the grazing value of grassland. The number represented the weight use in the mean aggregation; (B) Representation of the transformation used for the input variable of the grazing indicators.

biomass and the vegetation height were proportional, and we made also an estimation of the biomass every two weeks using the height of the vegetation.

To assess the values of the different grassland uses, we used a multi-criteria analysis tool: TATALE (Tools for Assessments with Transformation and Aggregation using simple Logic and Expertise); for more information see http://umr-selmet.cirad.fr/en/products-and-services/proposed-products/tatale. The first step of TATALE entails converting all variables into scores ranging from 0 to 1. These scores are then aggregated in the second step. Different methods can be used to aggregate these scores. The scores can be aggregated by a weighted average (the expert can put different weight on the different scores). The other methods to aggregate are to choose one of the scores either the highest score (maximal) or the lowest scores. The aggregation can be successive. Some of the score are first aggregated to create new scores. These new scores are then aggregated together and so on until a single synthetic score is obtained. These processes are repeated for grazing, ensilage and hay. Elaboration of the indicators was discussed and achieved during a meeting with all the authors of the paper.

Results and discussion

Figure 1a presents the aggregation tree for the grazing value. This shows how we aggregated the different scores transformed from the measured variables (in black) into the different synthetic scores (in grey). The grazing value relies on the quality and the quantity of forage. For the quantity, we supposed that not all the standing biomass could be grazed, either because the grass was too short or too tall or because some species were unpalatable. Figure 1b presents the transformation used for the different inputs variables (in x axis the value of the inputs variables and in y axis the scores attributed). Similar aggregations were done for the ensilage and the hay. Table 1 presents scores for the grazing, ensilage and hay values of four mixtures at various times of season.

This example shows how the values of the different utilizations vary during the season. The grazing values of the different mixtures decreases with the season. However, the decrease is higher for the mixes 1 and 4 than the mix 2 and 3. The ensilage score is higher in middle of April for the MIX 2 and higher in the beginning of May for the mix 1, 3 and 4. For the hay, the highest scores were for the four mixtures in the middle of April. This work proposes an indicator for estimating the value of grassland from a battery of many criteria. This indicator could be used to compared different types of grassland and be used as decision making tools especially to choose adapted mixtures for the implantation of grassland. The TATALE method relatively easily creates complex indicators for research and expertise and provides a possible answer to the problem of selection variables in multi-criteria analysis.

Table 1. Scores for grazing (G), ensilage (E) and hay (H) for four different mixtures at 4 different dates. (0 means the worst value and 1 the best possible value).

Mixture	MIX1			MIX2			MIX3			MIX4		
Use	G	E	H	G	E	H	G	E	H	G	E	H
Beginning March	0.36	0.15	0.31	0.37	0.48	0.31	0.44	0.51	0.36	0.43	0.53	0.38
Middle March	0.28	0.49	0.28	0.28	0.52	0.30	0.33	0.61	0.39	0.28	0.58	0.36
Middle April	0.22	0.45	0.47	0.34	0.78	0.51	0.35	0.47	0.53	0.29	0.45	0.50
Beginning May	0.18	0.63	0.43	0.24	0.64	0.45	0.25	0.67	0.48	0.17	0.65	0.44

Effects of inoculant on the fermentation, microbial composition and aerobic stability of whole crop maize ensiled in big tube

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Abstract

The present study aimed to investigate the effect of inoculant, containing *Lactobacillus buchneri* in combination with homofermentative lactic acid bacteria on fermentation, microbial composition and aerobic stability of the whole crop maize ensiled under field conditions. Whole crop maize was harvested in the dough stage of maturity of grain (at 34% DM of the plant) and ensiled into hermetic plastic big tubes either without additives or inoculated with mixture of lactic acid bacteria strains *Lactobacillus plantarum*, *Entrococcus faecium* and *Lactobacillus buchneri*. The final application rate was 1.0×10^5 cfu g^{-1} forage. Ten replications (big tubes, 1,700-1,800 kg each) were used per treatment. The SAS statistical package was used to analyze the data. Inoculant significantly reduced pH value and significantly increased concentration of acetate. Moulds and yeasts numbers and surface area of the big tubes covered with the moulds were reduced and aerobic stability was improved in the inoculated silage compared to the control silage. Quantities of silage suitable for feeding after 35 days aerobic exposure were significantly higher for the inoculated than the untreated silage.

Keywords: aerobic stability, inoculant, maize, silage

Introduction

Well fermented whole crop maize silage is an important source of dietary energy for ruminants due to the high energy content, but it is often prone to aerobic spoilage. Feeds that have undergone aerobic deterioration, may result in aerobic growth of yeasts and fungi, reduced nutritional value and present hazards to the animals and environment. Lactic acid bacteria inoculants were topical during the last two decades and their potential to improve silage fermentation was widely demonstrated (Muck, 2012). Combining heterofermentative with homofermentative LAB allows active fermentation and gain positive attributes when silages are exposed to air, respectively (Schmidt and Kung, 2010). The current study was designed to examine the effect of silage additive containing homofermentative LAB strains *Lactobacillus plantarum*, *Entrococcus faecium* and *Lactobacillus buchneri* on fermentation, DM loss, aerobic stability and microbial population of whole crop maize ensiled into a hermetic plastic big tube.

Materials and methods

Maize (*Zea mays*), was harvested and chopped in the dough stage of maturity of grain, 34% dry matter (DM) of whole plant with a maize harvester 'CLASS JAGUAR 840' adjusted to achieve a 10 mm theoretical chop length. Immediately after harvesting, the chopped forage was carted from the field to the ensiling and storage area and was ensiled into a hermetic plastic big tube (1.700-1,800 kg each), using Murska tube bagging equipment with a loading tunnel can and additive applicator. The following treatments were applied to fresh forage: control no additive and inoculated with homofermentative and heterofermentative LAB inoculant containing *Lactobacillus plantarum: Entrococcus faecium*: *Lactobacillus buchneri* – 20:30:50 (Chr. Hansen, Horsholm, Denmark; the stated total number of bacteria was 1.3×10^{11} colony forming units g^{-1} (cfu g^{-1}). The inoculant was applied at rate of 4 liter suspension per tonne forage according to the label instructions for application targeting a dosage of 1.5×10^5 cfu g^{-1} of fresh forage. To compensate for the water that was added to the treated silage, the control treatment (C) was sprayed with 4 l of tap water over a ton of fresh material to keep it at the same level of moisture as the treated silages. During the filling of the plastic big tube, one control bag (made from four layers cheesecloth) filled with 1 kg ensiling mass was inserted into each big tube for determining DM losses. Ten hermetic plastic big tube silages for each treatment were prepared. From each tube and treatment a core sample was taken at day 156 of storage for chemical and microbial analyses and for aerobic stability test. After storage period of 156 days, the big tube silages were subjected to an aerobic stability test at field conditions. The SAS statistical package was used to analyze the data. Silos were analyzed as a randomized complete block.

Results and discussion

Based on the dry matter content (339.6 g kg^{-1}), water soluble carbohydrates (88.3 g kg^{-1} DM) and crude protein concentrations (98.7 g kg^{-1} DM), and buffer capacity (20.6 mEq 100 g^{-1} DM) the whole plant maize was considered to be moderately easy to ensile. Inoculation accelerated fermentation, as identified by the lower ($P<0.05$) pH at day 2 after ensiling compared with the untreated control. Lower pH of the inoculant treated silages remained until day 156 of the storage (Table 1).

Inoculation with the blend of bacterial strains resulted in silages with higher ($P<0.05$) concentrations of acetic acid and lower lactic-to-acetic acid ratios. These results suggest that the fermentation in the inoculated silage was dominated by heterolactic *Lactobacillus buchnery* bacteria (Arriola *et al.*, 2011). Inoculated silages had lower numbers of yeasts and moulds at the time of removing of the plastic cover (day156 of the storage) compared with the control silages (Figure 1). However, during aerobic exposure

Table 1. Fermentation characteristics of big tube whole crop maize silages after 156 days of storage (n=10).

Parameters	Control	Inoculated	SEM	*P*
DM losses, g kg^{-1} DM	87	74	3.057	0.05
DM corrected for volatiles, g kg^{-1}	329	334	1.031	0.05
pH day 2	4.00	3.65	0.042	0.05
pH day 156	3.73	3.60	0.022	0.05
Lactic acid, g kg^{-1} DM	33.4	36.5	0.906	ns
Acetic acid, g kg^{-1} DM	17.1	20.3	0.532	0.05
Butyric acid, g kg^{-1} DM	0.50	0.30	0.048	0.05
Ethanol, g kg^{-1} DM	5.80	5.30	0.330	ns
Ammonia N, g kg^{-1} total N	32.7	31.0	1.141	ns
Aerobic stability, hours	100	168	7.853	0.05

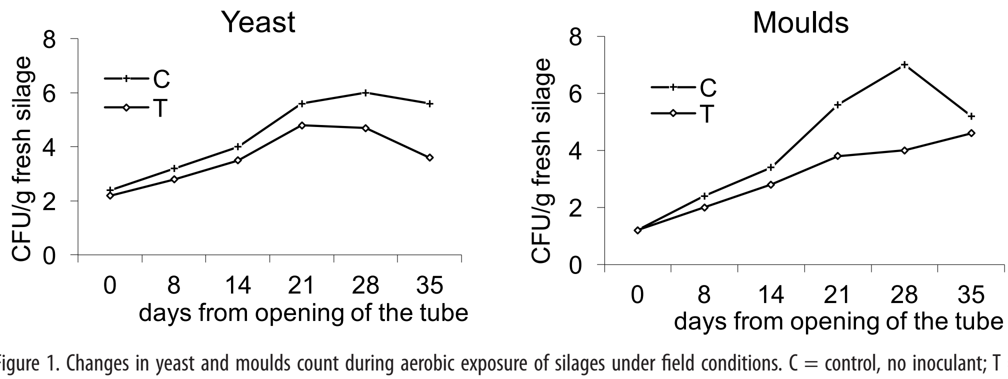

Figure 1. Changes in yeast and moulds count during aerobic exposure of silages under field conditions. C = control, no inoculant; T = *Lactobacillus plantarum*, *Entrococcus faecium* and *Lactobacillus buchneri*.

(at day 8, 14, 21, 28 and 35 after removing of the plastic cover) the number of yeasts and moulds increased more rapidly in the control silages than in the inoculated ones. The highest differences of yeast and mould counts between the control and inoculated silages were found at day 28 of aerobic exposure (5.99 vs 4.72 $\log_{10}$ cfu g^{-1} for the yeasts and 6.95 vs 3.99 $\log_{10}$ cfu g^{-1} for the moulds), which indicated that yeasts and moulds were effectively inhibited by the inoculant. Ström *et al.* (2002) showed the ability of a *Lactobacillus plantarum* strain to inhibit or change the morphology of selected fungi, due to antifungal cyclic dipeptides. Mari *et al.* (2009) indicated that the numbers of yeasts were lower in the silage treated with an inoculant versus the silage without inoculants. Inoculated silages had more hours to reach temperature more than 3 °C above the ambient, therefore, the aerobic stability of inoculated silages, on average, was greater than 68 hours compared with the control silages. Quantities of silage suitable for feeding after 35 days aerobic exposure were significantly higher for the inoculated than the untreated silage.

Conclusions

Inoculant containing *Lactobacillus buchnery* in combination with *Lactobacillus plantarum* and *Entrococcus faecium* gave more heterolactic type of fermentation, produce more acetate, reduced the number of yeast and moulds and improved aerobic stability.

Acknowledgements

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Ensilability of different mixtures of legumes and grasses

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Abstract

In Switzerland, mixtures of grasses and white or red clover are sown for leys. However some other legumes, such as sainfoin, contain bioactive secondary metabolites like condensed tannins which can improve health, production efficiency and product quality in ruminants. In 2015 a trial was carried out to investigate the ensilability and the silage quality of eight different mixtures from the first and third growth. In addition to different grasses, two mixtures contained red clover or red clover as well as lucerne and six mixtures only sainfoin as legume. The forage was pre-wilted to 410 and 356 g kg^{-1} dry matter (DM) for the first and third growth, respectively, and ensiled in laboratory silos. Concerning the chemical composition and the fermentability coefficient of the forage at ensiling, differences were found between the two different growths and also between some mixtures. But there was no direct influence of sainfoin visible. No problems with butyric acid were observed and all silages showed a good to very good silage quality.

Keywords: grass/legume mixtures, sainfoin, silage quality

Introduction

Legumes offer important opportunities for sustainable grassland-based livestock production systems (Lüscher *et al.*, 2014). However, legumes are considered more difficult to ensile due to low contents of sugars and high buffering capacity (Pahlow *et al.*, 2001). In Switzerland, the mixtures for leys contain besides different grasses also white or red clover or both. Other legumes, such as sainfoin (*Onobrychis viciifolia)*, contain bioactive secondary metabolites like condensed tannins which can improve protein utilization in ruminant livestock and have the potential for combatting parasitic nematodes in ruminants (Lüscher *et al.*, 2014). Sainfoin is used in grass/clover mixtures, which are less intensively managed. The ensilability and the silage quality of different mixtures with legumes and grasses were investigated in a field experiment.

Materials and methods

n 2015 a trial with eight different mixtures of legumes and grasses was carried out. The mixtures were sown in 2013 as part of the testing program of Agroscope in Changins, Switzerland, in small plots with four replicates. In addition to different grasses, two mixtures contained red clover as well as lucerne and six mixtures only sainfoin (Table 1). The forage was cut four times a year. For the silage experiment, the forage of three replicates of the first and third growth was used. The forage was pre-wilted to attain DM contents of 350 to 400 g kg^{-1}, chopped and ensiled in laboratory silos each having a volume of 1.5 l. The silos were stored at room temperature (approx. 20 °C). Chemical parameters were analysed by NIRS (NIRFlex N-500 FT-NIR Büchi Switzerland) before ensiling and after a storage period of three months. Fermentation acids, ethanol, ammonia and pH were also analysed in the silage. Furthermore, the fermentability coefficient of the fresh material was calculated. This parameter summarizes the potential effects of dry matter (DM) as well as the ratio of sugar content and buffering capacity on the fermentation. The botanical composition of the different plots was assessed visually. Data were analysed using analysis of variance and Bonferroni-Test (Systat 13).

Table 1. Seed composition of the eight different mixtures (kg ha^{-1})

Mixtures	1	2	3	4	5	6	7	8
Trifolium pratense	2	5						
Medicago sativa	15							
Onobrychis viciifolia			100	100	100	100	100	100
Dactylis glomerata	6	5	5	12				5
Arrhenatherum elatius		10	10		20			
Festuca arundinacea						18		15
Festuca pratensis	12	10					18	
Phleum pratense	3		3					3

Results and discussion

The proportion of legumes varied between 42 and 80% for the first growth and between 25 and 83% for the third growth (Table 2 and 3). The forage of the first and the third growth had an average DM-content of 410 and 356 g kg^{-1}, respectively. Among the eight mixtures, there were significant differences concerning the DM- and nutrient contents of the first and the third growth, but there was no direct influence of the legume sainfoin visible.

All fresh mixtures of the first growth and most of the third growth had fermentability coefficients over 45. A value above 45 indicates good fermentability (Pahlow *et al.*, 2001). Also the investigations of the Legsil project (Pahlow *et al.*, 2001) showed, that wilting legumes to DM-contents about 400 g kg^{-1} is

Table 2. Proportion of legumes, DM, nutrient contents and silage parameters of the eight different mixtures of the first growth (g kg^{-1} DM).[1,2]

	1	2	3	4	5	6	7	8	SE	*P*-value
Green forage										
Legumes, %	80^{a}	52^{bc}	57^{bc}	60^{abc}	42^{c}	67^{ab}	65^{ab}	65^{ab}	4.1	<0.001
DM-content, g kg^{-1}	434	392	384	422	417	408	368	452	17.4	0.059
Ash	95^{a}	80^{ab}	77^{ab}	81^{ab}	72^{b}	80^{ab}	78^{ab}	81^{ab}	3.4	0.013
Crude protein	177^{a}	135^{b}	134^{b}	128^{b}	122^{b}	131^{b}	136^{b}	128^{b}	7.6	0.003
ADF	317	359	338	332	344	323	326	329	9.2	0.102
NDF	470	541	492	512	524	471	476	506	19.3	0.149
Sugar	71^{b}	99^{a}	98^{a}	102^{a}	100^{a}	113^{a}	105^{a}	106^{a}	3.1	<0.001
FC	51^{b}	53^{b}	53^{ab}	58^{ab}	58^{ab}	58^{ab}	53^{b}	62^{a}	1.8	0.004
Silage										
DM-content, g kg^{-1}	428	384	386	425	418	405	366	450	18.2	0.078
pH	5,5	5,2	5,2	5,2	5,2	5,4	4,9	5,5	0.1	0.065
Lactic acid	19	32	24	19	24	17	35	17	4.5	0.103
Acetic acid	6	7	2	3	4	2	6	2	1.1	0.020
Butyric acid	1	2	4	2	2	3	2	1	0.9	0.346
Ethanol	5^{b}	12^{ab}	16^{ab}	18^{ab}	22^{a}	15^{ab}	22^{a}	14^{ab}	3.1	0,024
NH_3-N of tot. N, %	6,7	7,2	6,1	4,2	7,1	6,0	6,0	5,7	0.6	0.059
DLG points	90	90	78	85	89	85	90	90	2.7	0.111

[1] Mixture 1: red clover, lucerne and different grasses; Mixture 2: red clover and different grasses; Mixtures 3 to 8: sainfoin and different grasses; SE: Standard error, FC: fermentability coefficient; NH_3-N, % tot. N: ammonia-N as a proportion of total N.

[2] In the rows, superscript letters indicate differences significant at P<0.05.

Table 3. Proportion of legumes, DM, nutrient contents and silage parameters of the eight different mixtures of the third growth (g kg^{-1} DM).[1,2]

	1	2	3	4	5	6	7	8	SE	*P*-value
Green forage										
Legumes, %	83^{a}	25^{c}	35^{c}	30^{c}	42^{bc}	35^{c}	60^{b}	33^{c}	4.0	<0.001
DM-content, g kg^{-1}	363^{a}	383^{a}	329^{b}	343^{a}	332^{b}	370^{a}	351^{a}	374^{a}	9.3	0.006
Ash	100	114	108	114	106	97	94	104	4.5	0.045
Crude protein	206^{a}	204^{a}	188^{abc}	188^{abc}	206^{a}	167^{bc}	203^{ab}	161^{c}	5.2	<0.001
ADF	266^{a}	256^{a}	258^{a}	267^{a}	230^{bc}	253^{ab}	226^{c}	247^{abc}	4.7	<0.001
NDF	419^{abc}	465^{ab}	422^{abc}	469^{a}	395^{bc}	469^{a}	363^{c}	477^{a}	13.2	<0.001
Sugar	78^{d}	91^{bcd}	101^{abcd}	86^{cd}	102^{abcd}	115^{ab}	127^{a}	111^{abc}	5.1	<0.001
FC	45^{b}	49^{ab}	46^{b}	46^{b}	47^{b}	54^{a}	53^{a}	54^{a}	1.2	<0.001
Silage										
DM-content, g kg^{-1}	363^{ab}	379^{a}	329^{b}	341^{ab}	334^{ab}	371^{ab}	349^{ab}	371^{ab}	9.3	0.009
pH	$4{,}8^{b}$	$5{,}0^{ab}$	$4{,}8^{b}$	$4{,}9^{ab}$	$5{,}0^{ab}$	$5{,}2^{ab}$	$5{,}2^{ab}$	$5{,}4^{a}$	0.1	0.003
Lactic acid	71^{a}	35^{bc}	49^{ab}	39^{bc}	43^{bc}	24^{bc}	24^{bc}	19^{c}	5.0	<0.001
Acetic acid	26^{a}	9^{b}	12^{b}	6^{b}	10^{b}	5^{b}	5^{b}	4^{b}	2.2	<0.001
Butyric acid	0	1	5	3	4	1	1	1	1.6	0.496
Ethanol	11	7	12	8	12	8	7	6	1.3	0.017
NH_3-N of tot. N, %	$8{,}4^{a}$	$5{,}3^{bc}$	$6{,}6^{ab}$	$5{,}2^{bc}$	$6{,}6^{ab}$	$4{,}2^{c}$	$3{,}8^{c}$	$4{,}3^{c}$	0.4	<0.001
DLG points	92	90	78	82	82	90	90	90	6.9	0.773

[1] Mixture 1: red clover, lucerne and different grasses; Mixture 2: red clover and different grasses; Mixtures 3 to 8: sainfoin and different grasses; SE: Standard error, FC: fermentability coefficient; NH_3-N, % tot. N: ammonia-N as a proportion of total N.
[2] In the rows, superscript letters indicate differences significant at $P<0.05$.

necessary to obtain fermentability coefficients over 45. The fermentation parameters of the silages of the first and third growth are indicated in Table 2 and 3. In all mixtures, relatively high pH-values were found. The lactic acid production was inhibited. In all silages only very little butyric acid was produced and the ammonia proportion was in all mixtures lower than 10%.

All mixtures showed a good to a very good silage quality concerning the calculated DLG points (DLG 2006). No significant differences concerning the DLG points between the mixtures were found for the first, neither for the third growth.

Conclusions

The investigation showed that by wilting mixtures of legumes and grasses it is possible to increase the fermentability coefficients and to obtain silages of a good quality. The mixtures with sainfoin showed more or less the some nutrient contents and silage quality as the two mixtures without sainfoin.

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Effects of harvest delay on nutritive value in four grass and four legume species in binary mixtures

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Abstract

Information on companion species effects in grass-legume mixtures on quality changes during prolonged growth is lacking while this is relevant in view of delayed harvests that frequently occur in practice. Perennial ryegrass (*Lolium perenne*) was sown with either red clover (*Trifolium pratense*), white clover (*Trifolium repens*), lucerne (*Medicago sativa*) or birdsfoot trefoil (*Lotus corniculatus*), and white clover with hybrid ryegrass (*Lolium* × *boucheanum*), meadow fescue (*Festuca pratensis*) or timothy (*Phleum pratense*). Yield, concentrations of N, neutral detergent fibre (NDF), acid detergent fibre (ADF), lignin (ADL), and *in vitro* organic matter digestibility (IVOMD) were studied during two years in a cutting trial in Denmark. The dynamic development of compounds was investigated. Herbage digestibility and N concentration decreased during prolonged growth while contents of fibre compounds increased. In May, concentrations of ADF and lignin increased faster in legumes than in grasses; in August, NDF and ADF concentrations increased most quickly in legumes. Generally, N concentrations as well as IVOMD declined at similar rates in grasses and legumes but within each group, differences in yield and development of nutritive value occurred among species. The relationship between weekly growth rate and change in quality parameters differed among species and between functional groups.

Keywords: companion species, digestibility, fibre, functional group, harvest date, regrowth, yield

Introduction

In ruminant diets, herbage is an important natural source of protein and fibre. Knowledge about seasonal production patterns, relative contribution of each harvest to annual yield and nutritive value of individual harvests and total annual yield is needed for strategic planning of the grassland management in order to meet the herbage yield and quality requirements of the planned livestock production. Information on companion species effects in grass-legume mixtures on nutritive value changes during prolonged growth is lacking while this is relevant in view of delayed harvests that frequently occur in practice. Therefore a field experiment was conducted on a sandy soil with 7 two-species forage mixtures (Rasmussen *et al.*, 2012; Elgersma and Søegaard, 2015); annual yields and nutritive values were reported earlier. The aim of this experiment was to study changes in herbage yield and nutritive value of grasses and legumes in mixtures during prolonged regrowth in spring and summer. We hypothesised that relationships between weekly growth rate and weekly change in nutritive value parameters would differ among species and functional groups.

Materials and methods

Perennial ryegrass (*Lolium perenne* L.; 'PR') was sown with each of four forage legumes: red clover (*Trifolium pratense* L.; 'RC'), lucerne (*Medicago sativa* L.; 'LU'), and birdsfoot trefoil (*Lotus corniculatus* L.; 'BT'), and white clover (*Trifolium repens* L, 'WC') was sown with each of four companion grasses: perennial ryegrass, hybrid ryegrass (*Lolium boucheanum* Kunth; 'HR'), meadow fescue (*Festuca pratensis* Huds; 'MF') and timothy (*Phleum pratense* L.; 'TI'). Grass and mixtures were sown in 2006 in a small-plot (1.5×8 m) cutting trial with 4 replications in Denmark. Plots were fertilised to mimick farming practice with 300 kg N ha^{-1} from cattle slurry divided into four applications during the growing season of 100, 80, 60 and 60 kg and harvested five times in 2007 and four times in 2008 with a Haldrup forage harvester at a residual stubble height of 7 cm. Results of all nine harvests have been reported earlier (Elgersma and Søegaard, 2015). In May and August, the dynamic development of compounds and their interrelationships were investigated by sampling at the optimum harvest date ± one week. For analyses of hand-separated individual companion species, two composite samples were made: one of replicates 1 and 4, and one of replicates 2 and 3. Herbage yield, contents of N and neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL), and *in vitro* organic matter digestibility (IVOMD) were determined as reported by Rasmussen *et al.* (2012). In August, WC was separated in leaves and flowers. Within spring (May) and summer (August) harvests, species DM yield and concentration of compounds were analysed with a linear model with species, year and species × year as fixed factors and block as random factor (n=2). The weekly change was analysed by linear regression models of each parameter during 2-week intervals in May and August. Significance of differences in slope of the regression lines was tested among the species and between functional groups.

Results

Herbage digestibility and N content decreased during prolonged growth while NDF content increased. Grasses had a stronger yield increase than legumes in May (Table 1), although red clover grew as fast as the grasses. Within functional groups, the ranking order of species differed among seasons. While in May the growth rate of red clover was highest and that of birdsfoot trefoil lowest (Table 1), in August (not shown) red clover and lucerne had the highest, and white clover the lowest growth rate. In May, concentrations of ADF and lignin increased faster in legumes than in grasses; in August, NDF and ADF concentrations increased most quickly in legumes (not shown). Within the legumes functional group, large differences in growth rate and nutritive value changes occurred among species (Table 1).

Table 2 shows relationships between change rates of IVOMD, NDF and N contents per unit of DM. While in white clover the changes in nutritive value were not related to growth rate, birdsfoot trefoil showed the strongest decline in IVOMD and N content and increase in NDF content per unit growth. The decline in IVOMD and increase in NDF content per unit growth were stronger in lucerne than in red clover.

Table 1. Rates of change during 2-week growth intervals for dry matter yield (DMY), *in vitro* organic matter digestibility (IVOMD), concentrations of neutral detergent fibre (NDF), and nitrogen (N). Average values of 2 replicate plots during 2 years in May. Species abbreviations: red clover (RC), lucerne (LU), birdsfoot trefoil (BT), white clover (WC), perennial ryegrass (PR), timothy (TI), meadow fescue (MF), hybrid ryegrass (HR). S.E. is standard error.

Species	DMY (kg ha^{-1} w^{-1})	IVOMD (g kg OM^{-1} w^{-1})	NDF (g kg^{-1} DM w^{-1})	N (g kg^{-1} DM w^{-1})
RC	817^{ab} [1]	-7^{c}	23^{cd}	-3^{a}
LU	461^{c}	-37^{a}	38^{a}	-1^{b}
BT	45^{e}	-25^{a}	29^{ab}	-4^{a}
WC	240^{d}	-7^{c}	15^{e}	-4^{a}
PR	722^{b}	-16^{bc}	15de	-4^{a}
TI	697^{b}	-14^{c}	27^{bc}	-3^{a}
MF	697^{b}	-24^{ab}	31^{b}	-3^{a}
HR	905^{a}	-24^{ab}	33^{ab}	-3^{a}
Significance				
Species (S)	<0.0001	<0.0001	<0.0001	<0.0001
Year (Y)	NS	NS	<0.001	<0.0001
S ×Y	NS	NS	<0.0001	NS
S.E.	51.1	2.8	2.2	0.3

[1] Within a column, values without a common superscript are significantly different; *P*-values are shown.

Discussion

This study showed high growth rates in spring for grasses, particularly for hybrid ryegrass. The increased herbage NDF concentration with plant maturity is because accumulation of stem mass exceeds increment of leaf mass and therefore the leaf:stem ratio decreases. NDF concentrations are generally lower in legumes than in grass as was confirmed earlier in this field experiment (Elgersma and Søegaard, 2015), therefore legume content lowers total herbage NDF content in mixtures. Cell wall content is lower in leaves than in sheaths and stems, lower in legumes than grasses, and lower in young than old material. These group differences equate well with the inverse group differences in digestibility (Elgersma and Søegaard, 2015).

As hypothesized, relationships between weekly growth rate and change in nutritive value parameters differed among species and functional groups. However, as differences among legume species were much larger than among grass species (Table 1) we will focus on discussing relationships and implications among legumes. Within the legumes functional group, white clover had the lowest rate of change in concentrations of compounds and nutritive value during prolonged growth, making it robust against nutritive value decline in case of delayed harvest. This is particularly important in spring when the main silage cut is harvested for providing winter feed. Lucerne and red clover had comparable herbage production levels (Elgersma and Søegaard, 2015) and growth patterns (Table 1) in mixtures with perennial ryegrass, but red clover had a better herbage nutritive value, i.e. higher digestibility and protein content, and lower fibre compound concentrations than lucerne (Elgersma and Søegaard, 2015). Lucerne had a higher rate of change in IVOMD and NDF with time (Table 1) and per unit yield increase (Table 2) than the clovers, which implies that mixtures with lucerne would need to be harvested at the optimal moment to avoid loss in nutritive value during prolonged growth. Red clover was second best in terms of nutritive value (Elgersma and Søegaard, 2015). It was the highest yielding legume with the highest growth rate (Table 1), while changes in nutritive value per unit yield increase were relatively small (Table 2), indicating that red clover mixtures have benefits in practice.

High forage yields are needed for economical herbage production systems. With increasing milk yield per dairy cow, also the requirement for nutritive value of forage increases. Results of this study regarding

Table 2. Regression coefficients (*b*) and R^2 of linear relationships during 2-week growth intervals between *in vitro* organic matter digestibility (IVOMD) (g kg OM^{-1}), concentrations (g kg^{-1} DM) of neutral detergent fibre (NDF) or N (Y), and dry matter (DM) yield (X) ($Y = a + b^*$ t DM ha^{-1}) in herbage of four legumes and grasses in May. Based on 2 replicate plots during 2 years. Species abbreviations: red clover (RC), lucerne (LU), birdsfoot trefoil (BT), white clover (WC), perennial ryegrass (PR), timothy (TI), meadow fescue (MF), hybrid ryegrass (HR).

	IVOMD		NDF		N	
Legumes	***b***	**R^2**	***b***	**R^2**	***b***	**R^2**
RC	NS		20	0.71	-5.3	0.83
LU	-47	0.53	49	0.56	NS	
BT	-217	0.52	446	0.82	-35.1	0.52
WC	NS		NS		NS	
Grasses	***b***	**R^2**	***b***	**R^2**	***b***	**R^2**
PR	-11	0.42	NS		-2.5	0.41
TI	NS		NS		-2.3	0.63
MF	-17	0.69	25	0.80	-2.1	0.74
HR	-9	0.85	27	0.85	NS	

companion species effects on herbage yield and quality changes during regrowth in grass-legume mixtures can be used in practice to find a balance between these aims.

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The feed tables developed by INRA for forage quality evaluation: comparison with CVB and NorFor values

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Abstract

Forage feed tables developed at INRA in France are based on more than 2000 *in vivo* measurements of digestibility and voluntary intake in sheep accumulated over several decades with a standard protocol, and on measurements of *in sacco* nitrogen degradability (n=452). They cover a large variety of fresh forages, silages, and hays from grasslands and annual forage crop species. Feed value is expressed in net energy, in digestible protein in the intestine and in fill unit. An update of INRA feed tables will refine energy and protein values of forages and will include several novelties (e.g. fatty acid content of forages, new estimations of energy loss in urine and through methane emission). The relationship between digestibility, crude fibre and crude protein calculated on data from INRA feed tables are similar to those calculated on data from CVB (Netherland) and Norfor (Nordic countries) feed tables. This indicates good consistency between feed value databases from different European countries.

Keywords: feed value, forages, grasses, legumes

Introduction

Accurate evaluation of nutritive value and voluntary intake potential of forages produced from grasslands and annual forage crops is a key feature of their efficient utilization in diets. Tables of forage value exist in most countries and are regularly updated (e.g. INRA, 2007; CVB, 2011; NorFor, 2011). Generally speaking, forages are characterized in feed tables by: (1) their chemical composition; (2) *in vivo* or *in vitro* digestibility and N rumen degradability; and (3) energy and protein values calculated according the feed units of each country. Reference methods to measure digestibility and degradability vary between countries. For example, in France digestibility is measured on sheep fed *ad libitum* that allows also to measure voluntary intake and to estimate the intake potential of the forage expressed in fill units. This paper presents the French INRA tables of forage feed value and some novelties that are introduced in a new version. Finally, we compare the reference values provided by French tables (INRA) with those provided in Netherland (CVB) and in Nordic countries (Norfor).

Materials and methods

The main database that underlies the INRA tables contains more than 2000 *in vivo* measurements of digestibility and voluntary intake in sheep accumulated over several decades with a standard protocol. The database covers a large variety of grassland and annual forage crop species (3 types of permanent pastures, 8 grass species, 6 forage cereals species, 5 forage legume species, and 8 other species). A second database for *in sacco* N degradability (n=452) is used for estimation of protein value. The feed values of fresh forages are directly calculated from *in vivo* measurements and 294 forage types are categorized according botanical family, species, vegetation cycle, and stage of vegetation. According INRA (2007), energy value is expressed in net energy (1 UFL = 1,700 kcal), intake potential in fill units (1 LFU corresponds to a voluntary intake of 17 kg DM for a 600 kg cow producing 25 kg milk) and protein value in digestible protein in the intestine (PDI in g kg^{-1} DM). Feed value of conserved forages are estimated from the corresponding fresh forages using statistical models accounting for the modification of chemical composition, digestibility and voluntary intake induced by ensiling and making hay. Prediction equations of feed value components are established on feed tables and from enzymatic digestibility measurements (n=375). To compare INRA tabulated feed values with those from CVB and Norfor, we regressed the digestibility values on crude fibre and on crude protein content and tested (t test) if the slopes and the intercepts are different.

Results and discussion

The INRA tables account for the diversity of forages used in France: fresh and conserved forages used for different types of animals (dairy and beef cows, sheep, goats, growing animals). In addition to the feed values of fresh forages, feed values of 559 silages are provided according the type of conservation (direct cut or wilted silages, presence of an additive) and of 369 hays according the drying conditions (barn-dried, field-dried and the weather conditions). Figure 1 illustrates the variability of feed values of typical forages used to feed dairy cows. Fresh forages provide a high energy density and well balanced diet between protein and energy.

In a new version of INRA feed tables, energy, and protein values will take into account the effect of feeding level on digestion and an improved estimation of organic matter fermented in the rumen (Sauvant and Nozière 2016). Estimations of CH4 emissions, of N waste in the urine and of the related energy losses will be provided. Forage fatty acids content and fatty acids profiles will be characterized from the equations by Glasser *et al.* (2013). This will allow evaluating forage quality also in relation to other dimensions than nutritive value, as impact on product quality and emissions in the environment.

Plotting organic matter digestibility against crude fibre and crude protein content for INRA CVB and Norfor tables and the corresponding regression curves are shown in Figure 2. Digestibility decreases by 1.20, 1.24 and 1.23 g kg^{-1} DM for an increase of 1 g kg^{-1} DM crude fibre and increases by 0.71, 0.81 and 0.71 g kg^{-1} DM for an increase of 1 g kg^{-1} DM crude protein, respectively for INRA, CVB and Norfor tables. Neither the slopes nor the intercepts of the regression curves calculated on INRA, CVB and NorFor data were statistically different ($P>0.05$). This indicates a good consistency between the databases used to estimate forage feed values in France (INRA, 2007), Netherlands (CVB, 2011) and Nordic countries (Norfor, 2011).

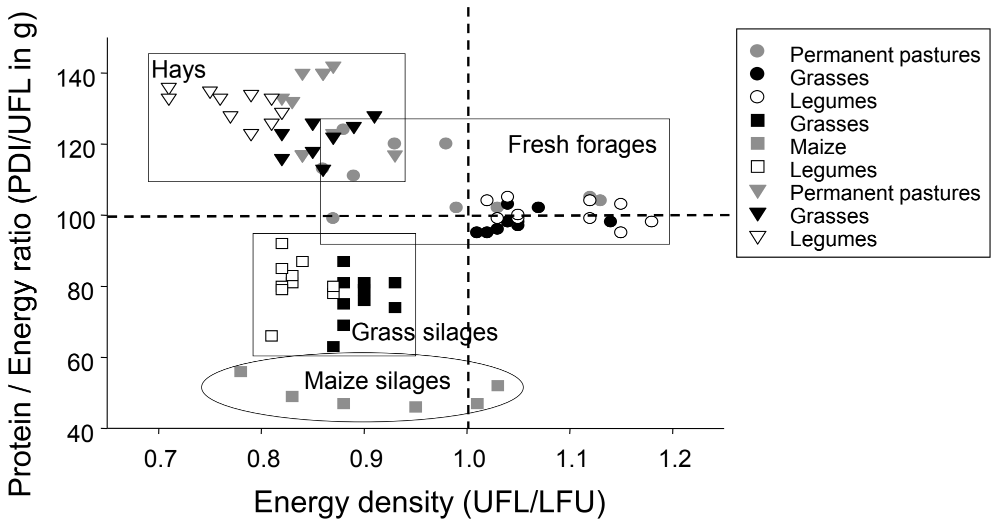

Figure 1. Variation of forage energy density and protein to energy ratio for typical forages used to feed dairy cows (data from INRA 2007).

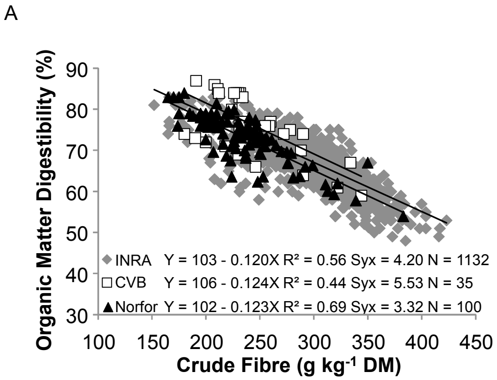

Figure 2. Relationship between organic matter digestibility and crude fibre (left), and between organic matter digestibility and crude protein (right) (data from INRA, 2007; CVB, 2011; NorFor, 2011).

Conclusions

INRA feed tables cover a large range of forages used in ruminant feeding. The reference data and the predictions equations are regularly updated to integrate advances of the research and to wider the scope of feed evaluation. The consistency of reference data used to estimate forage feed value in France, the Nordic countries and the Netherlands is promising for interoperability of databases and possible harmonization at a European level.

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Leaf:stem ratio as a tool to estimate field losses

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Abstract

Silage production from green forages often includes a pre-wilting in the field with a potential loss of plant material. The loss from the field has seldom been determined, but is expected to be high especially for legumes pre-wilted to high dry matter concentration. Under the assumption, that mainly leaf material is lost in the field, the change in leaf:stem ratio through the harvest management could be used as a tool to estimate field losses. To test this idea, the leaf:stem ratio was determined in samples collected before mowing, immediately after mowing, and after raking in fields with primary growth of pure perennial ryegrass (early and late harvested), festulolium, tall fescue and red clover. The estimated field losses were 8.7% and 2.3% in early and late harvested perennial ryegrass, respectively, 6.8% in festulolium, 24% in tall fescue and 12% in red clover. A high positive correlation (0.95) was observed between leaf:stem ratio in the initial sample and the estimated loss. It is concluded that leaf:stem ratio is a potential tool to estimate field losses.

Keywords: field loss, pre-wilting, forage, silage production, leaf:stem ratio

Introduction

In dairy farming it is important to have a high and stable forage production, and most green forages used for feeding are conserved as silage after a pre-wilting period in the field. In Denmark it is recommended to pre-wilt green forages to a dry matter (DM) concentration of approx. 350 g kg^{-1} as the in-silo DM losses due to effluent decrease with increasing DM concentration up to approx. 300 g kg^{-1} where it stop (Zimmer and Wilkins 1984). Concurrently, the pre-wilting period causes losses due to respiration, due to leaching by rain and due to mechanical treatment (McGechan, 1989). Field losses are expected to increase with increasing pre-wilting (Zimmer and Wilkins, 1984), especially for legumes pre-wilted to high DM concentration. Increased pre-wilting will reduce the microbial fermentation under the ensiling process, by which the protein value for dairy cows is increased (Johansen & Weisbjerg, 2015), but if field losses are high the total protein yield is reduced. The losses from the field during mowing, pre-wilting and raking have seldom been determined. At farm level it will be useful to have a tool to estimate field losses, which can be used to improve harvest management and consequently reduce field losses. Under the assumption, that mainly leaf material is lost in the field, the change in leaf:stem ratio in plant material collected in different steps of the harvest management can be used as a tool to estimate the field losses, and the objective of this paper is to test the practicability of this idea.

Materials and methods

Fields with pure perennial ryegrass (*Lolium perenne* L., cv. Calvano 1), festulolium (*Festulolium braunii* K.A cv. Perun), tall fescue (*Festuca arundinacea* Schreb., cv. Tower) and red clover (*Trifolium pratense* L., cv. Suez) were established in the start of April 2014 with barley (*Hordeum vulgare* L.) as cover crop on AU Foulum, Aarhus University, Tjele, Denmark. The primary growth of tall fescue, festulolium and part of the perennial ryegrass (early perennial ryegrass) were mown on May 21, 2015, and wilted for three days. In the middle of the wilting period it was raining 3.2 mm. The rest of the primary growth of perennial ryegrass (late perennial ryegrass) and the primary growth of red clover were mown on June 3, 2015, and wilted for two days. The goal with the wilting was to achieve a DM concentration of 350-400 g kg^{-1}. For all cuts the stubble height was set to 7 cm. The crops were wilted on broad swaths covering the whole area. After wilting, the swaths were raked before baling.

Five spots (30×30 cm), randomly selected in each field, were cut to 7 cm stubble height with a shears and pooled within species prior to mowing. Further, one sample covering 40 cm length of the swath width of a random selected swath was collected immediately after mowing and after raking in each field. All samples were representative reduced to a size of approx. 200 g and leaves (leaf blade and petiole) and stems (leaf sheath, stem and flower) were separated by hand and dried at 60 °C for 48 h to determine the leaf:stem ratio on DM basis. The field losses of DM were calculated with the assumption, that only leaf material is lost, whereas the stem quantity is constant. The following equation giving the loss in percentages was used:

$$\text{Field loss of DM (\%)} = (\text{ratio}_1 - \text{ratio}_2) / (\text{ratio}_1 + 1) \times 100$$

where ratio_1 is the first achieved (without loss) leaf:stem ratio and ratio_2 is the second achieved (with loss) leaf:stem ratio. No statistics were made, as the objective was to test the practicability of an idea.

Results and discussion

The leaf:stem ratio varied before mowing from 0.60 in late perennial ryegrass to 4.07 in tall fescue (Table 1). For festulolium the ratio after mowing was higher than the ratio before mowing, and for late perennial ryegrass and tall fescue the ratio after mowing was lower than the ratio after raking, by which negative field losses appeared (Table 2) when estimating the loss in the individual steps. To use the leaf:stem ratio as a tool to estimate field losses it is important, that the sampling procedure is performed well and is representative. The experience from this study indicates that sampling in the broad swath after mowing was difficult, whereas it was easier to get a representative sample from the narrow swath after raking. Therefore, the most probable estimates for the field losses in the grasses were the total losses from before mowing to after raking, which varied from 2.3% in late perennial ryegrass to 24% in tall fescue (Table 2). For red clover, the leaf:stem ratio before mowing was lower than the leaf:stem ratios after

Table 1. Leaf:stem ratio and the dry matter concentration (DM, g kg^{-1}) for the different crops before mowing, after mowing and after raking.

	Before mowing		After mowing		After raking	
	Ratio	DM	Ratio	DM	Ratio	DM
Early perennial ryegrass	1.30	199	1.21	211	1.10	346
Late perennial ryegrass	0.60	202	0.52	206	0.56	372
Festulolium	0.83	163	1.05	169	0.71	283
Tall fescue	4.07	194	2.31	200	2.86	412
Red clover	0.74	124	1.15	117	0.89	335

Table 2. Estimated field losses of dry matter (%). Suggested most probable estimates of total field loss are marked with an asterisk.

	Before mowing to after mowing	After mowing to after raking	Before moving to after raking
Early perennial ryegrass	3.9	5.0	8.7*
Late perennial ryegrass	4.8	-2.6	2.3*
Festulolium	-12	17	6.8*
Tall fescue	35	-17	24*
Red clover	-23	12*	-8.3

mowing and after raking. This was probably because the stubble height at 7 cm was not achieved during the mowing, since the red clover has lain down in the field before mowing. Therefore, the leaf:stem ratio after mowing and after raking cannot be compared with the leaf:stem ratio before mowing for red clover in this experiment, which was obvious from the estimated negative field losses. This also demonstrates, that the leaf:stem ratio as a tool to estimate field losses only is valid, if the stubble heights are identical for both samples used for the estimation. Because of the discrepancy in stubble height, the best estimate for the field loss in red clover in this experiment was probably the estimate from after mowing to after raking, which was 12% (Table 2).

The estimated field losses were highly correlated (0.95) with the leaf:stem ratio in the initial sample, indicating that more leaves enhance the risk for field losses. This does also support the assumption, that mainly leaf material is lost in the field, as the estimated total losses will be even higher if stem material is lost as well. As the forage parts dries they become more susceptible to be lost in the field (McGechan, 1989), therefore the final DM concentration in the forage is expected to highly influence the loss. The achieved DM concentrations in the forages were positively correlated (0.57) with the estimated loss, but not in same extent as leaf:stem ratio. Leaf material is drying faster than stem material, by which the DM concentration in the leaf part were appreciably higher in the leaf part compared to the stem part after wilting (data not shown), which increases the risk for losing leaf material.

The leaf:stem ratio as a tool to estimate field losses can only be used in fields, where both leaf and stem material is harvested. In the present experiment a field with pure white clover (*Trifolium repens* L.) was established as well, but due to the stoloniferous growth of white clover, only leaves were harvested in the vegetative phase. A high field loss was expected in this field, but the tool could not be used to detect the loss. Alternatively, the tool possible can be used in a modified way using the leaf blade:petiole ratio, but this idea has not been tested. Contrarily, the tool can be used in mixed grass-white clover fields, as the grass contributes with a stem part.

Conclusions

Leaf:stem ratio is a potential tool to estimate field losses for forages that have both a stem part and a leaf part. The tool requires representative sampling and a constant stubble height in the samples used for the estimation.

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Effects of heifers and sheep grazing on herbage production on a previously abandoned grassland

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Abstract

Large areas of cultivated grassland are annually abandoned and no longer used for production in Norway. Such areas will over time be encroached by shrubs and trees, which is regarded as undesirable. We assessed plant community development, pasture production, herbage quality and pasture utilization by sheep and heifers of a grassland that has been unmanaged for 12 years. The experiment was run for two consecutive years. Sheep grazed the whole area for one month in spring and autumn. During the summer, the area was assigned to three replicated treatments: (1) control with no management; (2) grazing heifers; and (3) grazing sheep with offspring. The stocking rate was 1.8 LU ha^{-1}, in both b and c, for a duration of one month. The area was left resting for a month after treatment and before autumn sheep grazing. Pasture production and herbage intake was estimated using grazing exclosure cages. Herbage consumed during summer period was on average 211 g DM m^{-2} and the pasture utilization was 55%. The annual consumption and utilization was 336 g DM m^{-2} and 62% in the grazed treatments and 28 g DM m^{-2} and 15% in the control, respectively. Total annual pasture production was on average 72% higher in the grazed treatments compared to the control. There was no difference between the grazed treatments on annual production, herbage intake or pasture utilization. Grazing stimulated herbage production, and such abandoned grasslands are valuable forage resources.

Keywords: grassland, productivity, utilization, botanical composition

Introduction

In Central Norway, the total farmland area was reduced by 4.9% (11 100 ha) between 2002 and 2012, of which grassland accounted for 80% of the decrease (SSB 2016). The reasons for abandoning farmland varies. Some land is lost to roads and urbanization. High quality land is commonly preferred at the expense of more marginal, remote and less suitable areas for intensive farming. Societal reasons, such as land ownership, does also influence land use. The abandoned areas will over time be encroached by shrubs and trees. This is regarded as undesirable because grasslands are important cultural landscape elements and agricultural land is scarce. Access to grassland is a limiting factor in sheep production. The use of grazing areas in spring and autumn are common but at the expense of yield of winter feed. Further, a general change from small to larger sheep flocks implies that there is a need for access to land for those that invest in future sheep farming. We elaborated the grass and animal production potential of abandoned grassland and the societal constraints that obstruct sheep farmers from using or getting access to such areas. The objective of the work presented here was to test the effect of sheep and heifer grazing on herbage yield and utilization of previously abandoned grassland.

Materials and methods

A 15.3 ha grassland area that had been unmanaged for 12 years was selected for the study. Before abandonment, the area was used as pasture for dairy cows. The prevailing plant species prior to onset of the experiment, assessed autumn 2013, included species that most likely were cultivated previously, such as common bent (*Agrostis capillaris* L., 37% of dry matter (DM) yield), smooth meadow-grass (*Poa pratensis* L., 12%), and naturally occurring species such as tufted hair-grass (*Deschampsia cespitosa* (L.) P. Beauv., 18%) and meadow buttercup (*Ranunculus acris* L., 7%). Reed canary-grass (*Phalaris* *arundinacea* L.) constituted dense stands in small patches. The experiment was run for two consecutive years. The area was not fertilized, and the soil is of morainic origin with high content of organic matter (ignition loss 12.1%), with moderate pH (5.4) and P (P-AL=7.8 mg 100 g^{-1}) and K (K-AL=5.8 mg 100 g^{-1}) contents. Sheep grazed the whole area for approximately one month in spring (Period 1: from May 23/20 to June 20/19 in 2014/2015) and autumn (Period 4: from September 17/14 to October 21/14 in 2014/2015). During the summer (Period 2), the area was assigned to three treatments: (1) control with no management; (2) grazing heifers; and (3) grazing sheep with offspring. The fields were on average 1.7 ha, and each treatment had three replicates. The stocking rate was 1.8 LU ha^{-1}, in both b and c, for a duration of approximately one month (from June 20/19 to August 12/3 in 2014/2015). The area was left resting for about a month after treatment and before autumn sheep grazing (Period 3: from August 12/3 to September 17/14 in 2014/2015).

Pasture production and herbage intake were estimated using grazing exclosure cages. Five cages, size 2 m^2, were placed at random in each of the nine paddocks. The cages were moved after each period. Herbage was cut at ground level inside and outside the cages (0.25 m^2) at 4 occasions when spring grazing finished, after summer grazing, end resting period, and after the end grazing in October. The samples were sorted into edible species and non-edible species (meadow buttercup, marsh thistle (*Cirsium palustre* L.), and dead material), forced dried at 60 °C for 48 hours and weighed. Herbage consumed was calculated by subtracting the yield outside from the yield inside the cages, and herbage utilization was calculated as herbage consumed divided by yield inside the cages. Annual herbage production was calculated by adding the herbage production in each period. The herbage production in each period was calculated by subtracting the yield estimates outside the exclosure cages at the start of each period from the yield in the exclosure cages at end of each period. Herbage botanical composition was estimated by using the dry-weight-rank method (Jones and Hargreaves, 1963) and by estimating plant ground cover for individual species in four permanent plots of 1 m^2 in each field in autumn (2013, 2014, and 2015) and spring (2014, 2015). Total DM yield and pasture utilization were analysed by using the mixed procedure in SAS (SAS 2011) to discern significant effects of treatment. There were no significant effects of year or year by treatment interaction and these effects were therefore omitted as fixed effects from the analysis.

Results and discussion

During the spring period (period 1) the accumulated total and edible DM yields were on average 196 (SEM 25.6) and 136 (SEM 18.8) g m^{-2}, and the sheep consumed on average 47 (SEM 12.7) g m^{-2}. In the summer (period 2) when the area was subjected to the different grazing treatments, the sheep and heifers consumed on average 211 g DM m^{-2} and utilized about 55% of the total biomass (Table 1). There were no significant effects of animal species on herbage production, consumption and utilization. The additional grazing with heifers and sheep during summer (period 2), resulted in 72% more total biomass production (526 vs 306 g DM m^{-2}), 159% more biomass production of edible plants (323 vs 125 g m^{-2}), 12 times more biomass consumed (336 vs 28 g m^{-2}) and 4.2 times higher utilization of the total biomass produced (0.63 vs 0.15) on an annual basis than in the control plots (Table 1).

No effects of grazing system on plant community were observed during the experimental years. In October, common bent was the prevailing species in all paddocks (on average 50% of DM yield, SD=10.3), whereas in May meadow buttercup dominated (32, SD=6.8%). Meadow buttercup was more widespread than creeping buttercup (*Ranunculus repens* L., average 2, SD=1.8% in October). Proportions of tufted hair-grass varied greatly between fields, but the species was evenly distributed between the treatments and accounted for 17 (SD=6.6) and 18 (SD=9.5) % of DM in yield in spring and autumn, respectively. The proportion of smooth meadow-grass was higher in October (11, SD=7.8%) than in May (1.5, SD=1.69%). In October 2015 (57%), the ground cover of dead material was higher than in October 2014 (31%). In total more than 60 plant species were registered. Most likely, the short duration of the

Table 1. Herbage consumed and pasture utilization by sheep and heifers during the summer period (period 2) and annually of two consecutive seasons of a previously abandoned grassland (n=6).

	Treatment			SEM	*P*-value
	Control[1]	Heifers	Sheep		
Period 2					
Consumed, g m^{-2}	5	215	208	33.4	0.018
Utilization, proportion consumed of total	0.06	0.57	0.52	0.102	0.035
Annual, period 1 to 4					
Total yield, g m^{-2}	306	526	527	47.0	0.040
Edible yield, g m^{-2}	125	320	327	42.4	0.042
Edible, proportion of total	0.41	0.59	0.61	0.112	0.106
Consumed, g m^{-2}	28	348	323	46.4	0.012
Utilization, proportion consumed of total	0.15	0.64	0.61	0.094	0.009

[1] Control is plots without grazing during the summer period, period 2.

experiment explains why no effects of grazing treatments on botanical composition could be observed. The plant species present at the onset of the experiment were species that are adapted to grazing. Grazing of the entire area with sheep in spring and autumn, short grazing period during summer and variation of plant communities between fields contributed to veil potential differences between treatments. The interaction of grazing livestock and plants is well known (McNaughton, 1979; Matches, 1992), but these interactions are often complex and plant community succession may occur slowly. In addition to grazing, mechanical cutting, draining, fertilisation, liming and reseeding with pasture seed mixtures may help to improve yields and feed value of herbage from abandoned grasslands. In the studied grassland, focus should be on liming and removing dead stems and leaves of tufted hair-grass, marsh thistle and reed canary grass.

Conclusions

Grazing during summertime stimulated herbage productivity up to 1.7 times that on ungrazed control plots, but two years of grazing had no significant effect on plant community composition.

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Showcase Hardenberg (NL): effect of variable manure rate applications on grass yields

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Abstract

Evolving in the past decades, GPS techniques have changed agricultural management. Combining precise location with yield and soil maps, variable rate applications (of pesticides, fertilizers, seeds, etc.) 'on the spot' can improve yields and/or quality of crops. The concept of Smart Farming Technologies aims to integrate spatial information, such as yield maps, with local knowledge and strategy from the farmer in order to give a plant or soil the specific treatment it needs. In 2015 the lectureship 'Smart Farming' at CAH Vilentum carried out a grass field trial with yield maps generated by a forage harvester. The goal of this pilot-research was to investigate in which way variable rates of manure can help to improve yield and soil quality as a basis for a sustainable production of grass. Grass yield of swards with regular rate (FIX) and swards with variated rates (VAR) of manure were compared (rate based on previous yield). During each harvest, yield was measured with a sensor and dry matter content (DM) with NIR on the forage harvester and by taking samples of the grass. Samples were analysed on DM, feed quality and nitrogen content. Comparing FIX and VAR, local growing factors (soil type, moisture, rooting depth, micro nutrients) seem to be more important in varying manure rate than the previous yield. VAR based on previous yield showed a lower yield than FIX, meaning variation in manure rate should be based on more factors (soil conditions and farmers' knowledge).

Keywords: variable manure rate, manure management, smart farming grassland

Introduction

Over recent years GPS techniques have changed agricultural management. Next to executing applications on the fields more precisely, location based yield maps give more insight in the variations within a field. When these yield maps are combined with knowledge of the field (e.g. soil data, yield previous crops), variable rate applications might help optimise yields and quality (Wittry and Mallarino, 2002; Long *et al.*, 2016). The concept of Smart Farming Technologies aims to integrate spatial information, such as yield maps, with local knowledge and strategy from the farmer (e.g. feed composition) in order to give a plant the specific treatment it needs (Kempenaar and Kocks, 2013). Farmers are faced with the challenge to produce more feed for the dairy stock, but at the same time they have to reduce their nitrogen and phosphate usage (EU, 1991). This fact combined with the availability of yield maps, generated by sensors on forage harvesters, raised the question whether it is possible to apply variable rates of manure on grassland based on yield maps. The goal of this pilot-research was to investigate how the concept of Smart Farming Technologies can help to optimise the rate of return (in terms of yield) of grassland for a dairy farm, aiming at closing the nutrient cycle and improve the soil quality as a basis for a sustainable production of grass. This paper summarises and discusses results from the field trial comparing fixed and variable rate applications of manure on a sandy soil.

Materials and methods

The trial field, a grassland of eight hectares on a sandy soil, was subdivided into four strips of 26 m width (three swards each) and approximately 250 m length. Two strips were treated with a regular rate (FIX) and two strips with variable rates (VAR) of manure, making a total of four times three swards (Figure 1). The basic principle used for VAR was that the amount of applied nitrogen in the manure, should be

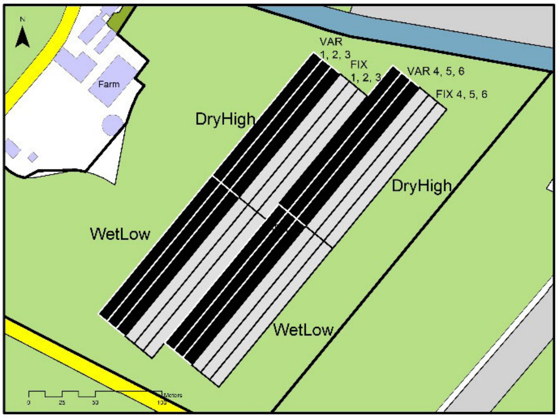

Figure 1. Trial field at Hardenberg.

the same as the amount harvested in the previous yield (in accordance with agricultural software like Farmworks, www.farmworks.com). Higher yield thus followed by more manure in order to maintain a constant and sustainable nutrient level in the soil. Previous to the first cut in May, the whole field received a fixed dose of 25 m^3 manure ha^{-1}. The other (three) applications during the growing season were either the FIX (15 m^3 manure ha^{-1}) or VAR (each time based on the yield variation of the previous cut, variation rates 3-30 m^3 manure ha^{-1} on the go, with a mean of 15 m^3).

Local growing conditions, in general, have a substantial influence on yield. For this reason, the soil was scanned in May with a sensor by Medusa Explorations (www.medusa-systems.com) and in all 50 locations in the field were described by physical analyses of soil samples and altitude. This showed that moisture was mainly related to altitude. Due to the fact that half of the field was lower in altitude, a split in the set up was made by introducing factor Location (WetLow and DryHigh), in order to investigate the influence of this effect.

During each cut, fresh yield was measured with a flow sensor and DM with NIR on a forage harvester (Digman and Shinners, 2008; Kumhála *et al.*, 2007; Shinners *et al.*, 2003). Yield data were analysed by analysis of variance (ANOVA) in a split-split-plot design using GenStat® Release 12.

Results and discussion

As expected, yield in May showed no differences between VAR and FIX (Table 1), since there was no difference in application. In June and July VAR resulted unexpectedly in a significant lower yield compared to FIX as the areas with prior high yields received a higher dose of manure. Explanations for this can be found in the local growing conditions such as soil type and moisture (Location). In August there was again no difference in VAR and FIX. This can be explained by the weather conditions (dry), overruling the influence of the differences in manure application.

Location (Table 2) varied in yield due to changing moisture conditions. In a wet period (May), the WetLow part had suboptimal growing conditions (too wet) while DryHigh had better conditions. Getting dryer in June, WetLow had the best conditions resulting in the highest yield. July and August were dry, turning too dry for both locations in August, resulting in a low yield. Due to the changing weather conditions application of more manure, to compensate higher nutrient abduction (higher yield) through VAR, did not result in higher yields in the subsequent cuts. Although this could be interpreted as a disappointing result, it leads to an important insight for the strategy being used for fertilising the field. Instead of varying the rate of manure based on the previous harvested product, the varying should be based on the expected yield and growing conditions. For example, a dry location in the field gave a higher grass yield in May, while

Table 1. Interaction between manure and harvest date in dry matter yield (tons ha^{-1}). [1]

Manure	Harvest date			
	May	June	July	August
FIX	2.9^{d}	4.4^{f}	2.2^{c}	1.2^{a}
VAR	3.1^{d}	3.7^{e}	1.8^{b}	1.2^{a}

[1] P<0.001; LSD=0.217; same letters = no differences.

Table 2. Interaction between location and harvest date in dry matter yield (tons ha^{-1}).[1]

Location	Harvest date			
	May	June	July	August
DryHigh	3.4^{e}	2.8^{d}	2.1^{c}	1.4^{b}
WetLow	2.7^{d}	5.3^{f}	1.9^{c}	1.1^{a}

[1] P<0.001; LSD=0.217; same letters = no differences.

the wet part produced better in June. In the current strategy, the dry spot received a high dose of manure after the yield in May. This created a high loss of nutrients since the plants cannot use this manure in the relative dry spring (nor in the dry summer) when moisture is limiting. Together with the yield maps this data has been discussed with the farmer to find potential correlations to explain the variations in the yield and hence to optimise the fertilisation strategy for the coming years. Knowledge of the field should be implemented in the strategy. During the year several discussions, based on the yield maps combined with soil maps, lead to a better understanding of the soil-plant system. The local knowledge of the farmer plays a key role in this discussion. This knowledge of the soil-plant system is essential since it might be more important to predict the future yield rather than knowing the harvested yield.

Conclusions

VAR had a lower production than FIX in interaction with the harvest date, so the variable rate based on the previous yield map was not sufficient to generate a comparable or higher yield.

Instead of varying the rate of manure based on the harvested product, we expect that the varying should be based on the expected yield. Also Location (growing conditions) showed interaction with the harvest date. This leads to the conclusion that knowledge of the growth factors in the field is essential in the process of making a profitable variable fertilisation strategy. With the knowledge of the growing season and the physical and chemical differences in the soil, the fertilisation strategy for the coming year will be adapted. Only locations that have the potential to use manure efficiently, will receive higher manure doses.

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Nutritive and fermentative quality and stability over time of silages made with winter forage legumes

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Abstract

Livestock profitability requires feeds of high quality and low cost. Therefore, adding legume silages in dairy cows diets are an interesting option. The CAP provides specific support for rotational cropping systems that include legumes and others protein sources in order to restrict protein imports and preserve the environment. The aim of this study was to compare the nutritive and fermentative characteristics and pH and temperature changes after air exposure of silages including legumes as compared with Italian ryegrass (*Lolium multiflorum* Lam. (IR)) silage. For this purpose three legumes (*Lupinus albus* L. (LA), *Vicia faba* L. (FB) and *Trifolium pratense* L. (TP)) grown in monoculture or in intercropped with IR or *Camelina sativa* L. (C) were evaluated in a completely randomized design with three replications. Laboratory silages were made with the harvested forages and were opened and analyzed after a fermentative period of 60 days. The protein contents were higher with legume silages alone than IR silages (P<0.001). The pH, N-NH_3, lactic and volatile fatty acids values obtained indicate that legume silages had an adequate fermentation, despite FB promoted higher proteolysis than other legumes ensiled either alone or in mixture. Legumes had better stability over time after air exposure compared with IR.

Keywords: protein self-sufficiency, silage quality, conservation

Introduction

The dependence on protein feed import is a weakness of EU agricultural systems. For this reason, the Common Agricultural Policy (CAP) provides specific support for rotational cropping systems that include legumes since they are good sources of protein with positive effects on both the environment and animal nutrition. The legumes' N fixation ability allow reduced inorganic N-fertilizer inputs (Lüscher *et al.*, 2014) and provide N for companion plants grown in association with legumes or for subsequent crops in the rotation (Rochon *et al.*, 2004). However, in comparison to grass, legumes are more susceptible to proteolysis in the silo due to their higher crude protein content, lower carbohydrate content and greater buffering capacity (Foster *et al.*, 2011). An alternative to cope with this problem is the use of grass-legume mixtures. Copani *et al.* (2014) showed that some bioactive legumes as mixtures with grass at the time of ensiling not only improve the fermentation process but also preserve silage quality via reduced protein degradation. Moreover, the energy: protein ratio in these mixtures increases biomass production by transferring the N symbiotically fixed from legumes to grasses and this can stimulate voluntary intake (Niderkorn *et al.*, 2015). The aim of this study was to compare the nutritive and fermentative values and the stability over time of legume silages in monoculture or in association with grasses against Italian ryegrass silage.

Materials and methods

Three legumes, *Lupinus albus* L. (LA), *Vicia faba* L. (FB) and *Trifolium pratense* L. (TP) growing in monoculture or associated with *Lolium multiflorum* Lam. (IR) or *Camelina sativa* L.-(C) were evaluated as winter crop alternatives to IR in a completely randomized design with three replications. After harvest in spring 2014, fresh herbage was ensiled at room temperature in laboratory silos during a fermentative period of 60 days. After that, all silos were opened and three sub-samples of each experimental silage were collected. The first one to determine pH, ammonia-N by distillation and lactic acid and volatile fatty acids by HPLC (Waters, Milford, MA) in aqueous extract. A second sub-sample was freeze-dried to avoid loss of volatile compounds and after that was milled at 0.75 mm and analysed for nutritive value (AOAC, 1984). The last sub-sample was used to observe pH and temperature changes over time during ten days after opening and exposing to air silages in a chamber with controlled temperature (20±1 °C). Nutritive and fermentative parameters were analysed as ANOVA (R Core Team, 2014) using species as main factor. The averages were compared against IR results that were used as control.

Results and discussion

Nutritive and fermentative characteristics of silages tested are presented in Table 1. As expected, the protein contents were higher in monoculture legume silages than IR silage (165, 166 and 168 vs 103 g kg^{-1} DM for TP, FB, LA and IR, respectively; $P<0.001$). The grass-legume mixtures, as FB+IR, FB+C and LA+C, also represent a good alternative for self-sufficient protein ($P<0.001$). LA, TP and C silages had high digestibility values and energy content without differences with IR silage ($P>0.05$). With regards to fermentative characteristics, in general, the legumes did not promote substantial changes on the amounts of lactic and acetic acids, while FB promotes higher proteolysis than other legumes either alone ($P<0.05$) or in association with other species (FB+IR and FB+C; $P<0.001$).

In Table 2 are shown the pH and temperature values during ten days after silos were opened. Taking into account the average room temperature during this period of exposure (20±1 °C), legumes and camelina (C) in monoculture and any grass-legume mixtures, as FB+IR or LA+IR, improved the stability of silages after air exposition when comparing with IR, since they maintained a stable pH over time and the increase of silage temperature were retarded.

Conclusions

All the forage legumes tested increased the protein concentration of silages. Legume silages had adequate fermentation and good stability over time, even though FB promoted higher proteolysis than other legumes ensiled either alone or in mixture.

Table 1. Nutritive and fermentative characteristics of silages tested. See Material and methods for explanation of treatments.[1]

Treatment	pH	CP	ADF	NDF	IVOMD	ME	NH_3	Lactic acid	Acetic acid
IR	4.05	103	265	431	80.3	11.4	5.6	81	27
TP	4.23	165***	326*	444	79.9	11.4	4.6	59	15
FB	4.05	166***	465***	576***	71.7**	10.3*	9.0*	93	31
LA	4.04	168***	391***	455	82.3	11.9	6.7	104	19
C	4.04	139*	293	418	75.9	11.0	3.2	34*	15
TP+IR	4.10	67*	319	473	72.3**	10.4*	4.1	56	21
FB+IR	4.17	155***	426***	592***	65***	9.2***	10.2***	89	31
LA+IR	4.13	93	350**	504**	70.8***	10.0**	5.4	77	24
TP+C	4.31	124	365***	499**	70.3***	10.2**	3.3	41	11**
FB+C	4.16	157***	445***	566***	66.4***	9.6***	11.0***	102	33
LA+C	4.00	154***	339**	472	72.9***	10.5*	5.3	86	15
s.e.	0.027	6.0	11.3	10.7	1.01	0.15	0.48	4.6	1.5

[1] CP = crude protein (g kg^{-1}DM); ADF and NDF: acid and neutral detergent fibre (g kg^{-1}DM); IVOMD: *in vivo* organic matter digestibility (%); ME: metabolizable energy (MJ kg^{-1} DM); NH_3: g ammonia-N 100 g^{-1} total N; Lactic and Acetic acids: g kg^{-1} DM. The differences against IR are showed as: *: $P<0.05$; **: $P<0.01$; ***: $P<0.001$; s.e.: standard error.

Table 2. pH and temperature values during air exposure of silages (see material and methods for explanation of treatments).

	Day	Treatment										
		IR	TP	FB	LA	C	TP+IR	FB+IR	LA+IR	TP+C	FB+C	LA+C
pH	1	4.05	4.11	4.06	3.97	3.93	4.11	4.13	4.06	4.38	4.16	3.99
	2	4.10	4.05	3.95	3.88	4.37	3.94	4.05	3.85	4.25	4.35	4.21
	3	4.03	3.81	4.03	4.01	4.21	3.82	4.16	4.02	4.24	4.13	3.84
	4	5.12	4.21	3.98	4.11	4.21	4.88	4.27	4.06	4.62	4.16	4.13
	5	5.78	4.20	4.08	3.94	4.38	5.84	4.20	4.04	4.71	4.22	4.10
	6	6.43	4.24	4.06	4.03	4.26	6.73	4.31	4.01	4.99	4.18	4.06
	7	6.71	4.31	4.06	4.04	4.23	7.38	4.32	4.01	5.28	4.17	4.02
	8	6.85	4.34	4.12	4.08	4.20	7.56	4.18	4.03	5.63	7.21	4.30
	9	7.09	4.35	4.20	4.42	4.23	7.89	4.38	4.16	5.55	7.67	5.23
	10	7.33	4.45	4.31	5.33	4.44	8.11	5.57	4.53	6.59	8.62	6.93
T (°C)	1	19.3	20.7	18.0	17.8	17.5	20.4	18.1	17.8	19.9	17.8	17.5
	2	19.8	20.7	18.1	17.7	18.0	21.0	17.9	17.8	20.3	18.2	17.9
	3	22.6	21.1	18.4	17.8	18.2	27.7	17.8	17.9	21.2	18.6	18.1
	4	26.1	21.5	17.7	17.9	18.2	31.3	17.9	17.9	22.5	19.1	18.7
	5	24.3	22.1	18.0	17.9	18.3	28.6	18.1	17.8	23.4	20.0	19.9
	6	23.1	22.2	18.4	18.4	18.2	27.5	18.6	18.1	24.2	21.2	21.9
	7	22.8	21.5	19.0	19.9	18.3	25.9	19.7	20.5	23.0	21.2	23.6
	8	23.5	21.1	19.5	21.3	18.1	24.4	20.9	23.3	21.9	22.0	24.2
	9	23.9	21.1	21.6	22.5	18.6	23.4	21.4	24.3	21.5	21.6	23.1
	10	23.0	21.1	22.7	21.8	19.0	22.8	21.2	24.1	21.9	19.7	21.4

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Influence of nitrogen fertilization on the crude protein fractions of grassland forage

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Abstract

Fertilization of permanent grassland ensures satisfactory dry matter DM yields in the long-term. But intensive fertilization may also influence crude protein (CP) quality in forages, which may decide about N-use efficiency in feeding ruminants. In a permanent grassland experiment (cultivated since 1966 in Gumpenstein, Austria) five treatments (1: control – no fertilization; 2: P and K; 3: P, K, 80 kg N; 4: as 2 but plus 120 kg N; 5: as 2 but plus 180 kg N; r=4 each) were sampled in the first cut 2014. Fresh forage samples were separated into grass, legumes and herbs, dried (58 °C) and milled (1 mm). Protein fractions were analysed according to CNCPS (Fraction A, B and C) in bulk samples. Legume proportion was highest at treatment 2, whereas grasses benefited most from increasing N fertilization with highest proportion at treatment 5. In all treatments CP concentrations were highest in legumes, as expected. CP fraction A increased in the bulk sample proportionally to the N fertilization level. Concluding, grasses seem to dominate the forage production in all treatments. However, high N fertilization may reduce CP quality in permanent grassland with well-defined botanical composition.

Keywords: permanent grassland, crude protein fractionation, long-term experiment

Introduction

The contribution of grassland for the protein supply of ruminants becomes more and more important. In this context, not only the crude protein (CP) content in grassland forage is relevant, but also the CP fractionation which provides detailed information about the CP quality for ruminants. CP quality in grassland also implies in a contribution of a more efficient N use as a high N-excretion causes problems for the N balance at the farm.

For permanent grassland regions, decisive strategies to obtain high N yields per hectare involve the optimum botanical composition and the balance between the ideal cutting frequency, nutritive value and dry matter (DM) yield (Pötsch, 1998). One important aspect in intensively used permanent grassland, with up to 5-6 cuts year^{-1}, is the forage containing high proportions of proteins with a fast degradation rate. A low proportion of easy fermentable carbohydrates like water soluble carbohydrates, combined with a high proportion of fast degraded protein results in high production of ammonia in the rumen, and the surplus N is consequently excreted in urine. Improving the protein quality of permanent grassland would therefore also enhance the N use efficiency of ruminants. In N-unfertilized temporary grassland, forage legumes determined the feed quality of binary mixtures with perennial ryegrass (Gierus *et al.*, 2011). Although N fertilization suppresses legumes facilitating the dominance of grasses, CP quality may decrease. The objective of the present study was to determine the CP fractionation in permanent grassland plots with different levels of N fertilization.

Materials and methods

The samples originated from the first cut in 2014 of a long-term grassland experiment, established already in 1966 at Gumpenstein, Austria. Consequently, the botanical composition of the experimental plots is very well adapted to the fertilization strategy. In total five treatments, each with four replicates, were

Table 1. Overview of results for the first cut 2014.

	Fertilization level					SEM	*P*-value
	1	2	3	4	5		
DM yield, Mg ha^{-1}	0.74^{d}	1.90^{c}	3.40^{b}	3.59^{b}	4.57^{a}	321	<0.01
N yield, kg ha^{-1}	12.3^{c}	33.6^{b}	52.3^{a}	51.3^{ab}	68.4^{a}	4.7	<0.01
ADF, g kg^{-1} DM	303.4^{c}	332.9^{bc}	363.9^{ab}	369.5^{a}	388.6^{a}	7.9	<0.01
Crude protein, g kg^{-1} DM[1]	101.3	101.8	96.1	89.5	93.7	1.8	0.23
fraction A, g kg^{-1} CP	122.6^{b}	123.3^{b}	162.1^{ab}	155.0^{ab}	169.8^{a}	6.4	0.03
fraction B, g kg^{-1} CP	866.3^{a}	866.0^{a}	826.5^{ab}	832.3^{ab}	819.5^{b}	6.4	0.03
fraction C, g kg^{-1} CP	11.0	10.7	11.4	12.4	10.7	0.3	0.37

[1] Crude protein fractionation according to Licitra *et al.* (1996).

selected and evaluated: Treatment 1: control, no fertilization; Treatment 2: P and K dynamic, i.e. the P and K fertilization was adjusted to the DM yield of the year before; Treatment 3: P and K dynamic plus 80 kg N ha^{-1}; Treatment 4: P and K dynamic plus 120 kg N ha^{-1}; Treatment 5: P and K dynamic plus 180 kg N ha^{-1}. Fertilization was realized with NH_4NO_3 (27% N), phosphate (25% P_2O_5) and potassium (40% K_2O). At the harvest date of the first cut, the botanical composition as well as the development stage was estimated visually. Swards were cut to 5 cm height and DM yield was determined after drying. A bulk sample was collected, as well as a representative manual separation in grasses, legumes, and herbs was performed. All samples were dried in hot-air cabinet at 58°C, milled in a Cyclotech mill to pass a 1 mm sieve and stored for further analyses. The CP fractionation was performed for the bulk sample. Fraction A was determined with a 10% tungstic acid solution (Licitra *et al.,* 1996) and N measured in the residue after filtration. The fraction A was calculated as difference between total N content and N content in residue. For determination of Fraction C, acid detergent fibre (ADF) was determined using a semi-automatic apparatus (Fibertech, Gerhardt, Germany) and N determined in ADF residue. The Fraction B was estimated by difference between total crude protein and Fraction A and C on DM basis. Data were statistically analyzed using the GLM procedure of SAS as a completely randomized block design, with the least significant difference procedure for mean comparison and probabilities being adjusted by Tukey-Kramer test.

Results and discussion

DM yield clearly corresponded to the expectations with regard to fertilization intensity (Table 1). N yields increased with the fertilization level, although they did not differ among treatments 3-5, probably as a consequence of dilution effects. The highest level was obtained in treatment 5 with 68 kg N-yield ha^{-1} (Table 1), which corresponded to the highest DM yield. The dominant grasses were *Dactylis glomerata* and *Trisetum flavescens* in treatments 2-5. For legumes, *Trifolium pratense* and *Trifolium repens* were mostly observed. *Leontodon hispidus* dominated among herbs. The botanical composition of the sward in the first cut confirmed the clear differentiation between grasses, legumes and herbs in dependence of the fertilization level, which was expected after almost 50 years fertilizer application (Figure 1). The proportion of legumes was highest in treatment 2 and declined with higher N-fertilization level, achieving less than 10% (on DM basis) in the other treatments. Legumes benefited from P and K fertilization mostly. The highest proportion for grasses, with values ranging up to 80%, was observed in treatment 5.

With regard to the CP fractionation, the proportion of fraction A (NPN-fraction) increased with N-fertilization level, achieving high values in treatment 5 (Table 1). Consequently, low content of fraction B was also observed in the same treatment. As fraction C was not affected by fertilization level,

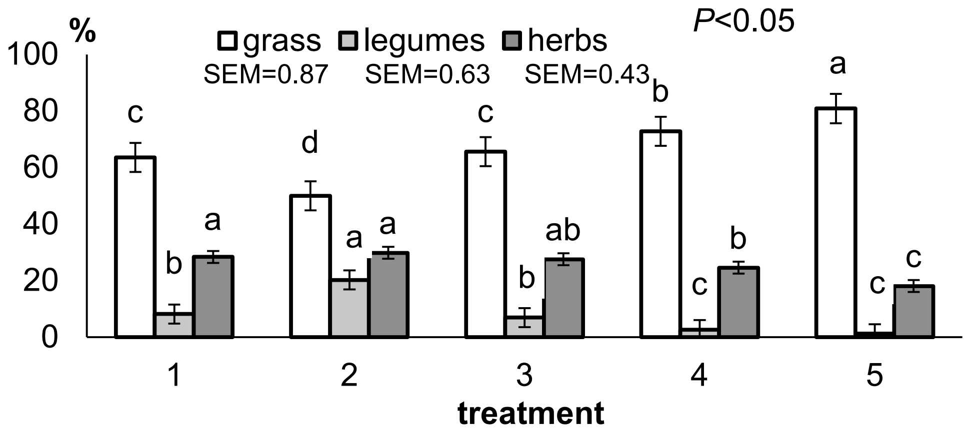

Figure 1. Botanical composition of experimental plot as averages (1993-2014).

the lower fraction B corresponded to the higher fraction A. Maybe the additional N uptake by the plant community in treatment 5 (mostly grasses) could not be converted into protein, and this N was retained as non-protein N in plant tissue. The CP quality suggests that lower N use efficiency in ruminant feeding is likely (Peyraud and Astigarraga, 1998).

As a long term fertilization trial, the cutting frequency is one of the major concerns. In practice, a higher fertilization level implies a higher cutting frequency. This was not performed in this experimental set up to avoid confounding effects over the years.

Conclusions

The knowledge about the botanical composition after almost 50 years of trial set up supports the observation that grasses are dominating with increasing level of N-fertilization. At the same time, the CP quality decreased. Although higher DM yields are possible with the highest fertilization level, the CP quality suggests that lower N use efficiency in ruminant feeding is likely. Moderate N fertilization supports higher legume proportion and adequate CP quality.

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Balanced phosphorus fertilization on grassland in a mixed grazing and mowing system; results after 18 years

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Abstract

In the Netherlands the P application standard including animal manure and P fertilizer has decreased to balanced P fertilization since 2015. Moving towards balanced P fertilization might affect grassland yield and quality, because soil processes might influence the P availability for plant uptake. In the short term, balanced P fertilization is not expected to limit dry matter (DM) production of grass on soils with a sufficient to high soil P. Decreases in P content and P offtake of grass are, however, expected directly after decreasing P fertilization. In the longer term decreases of herbage yield can be foreseen. With balanced P fertilization, annual DM yield and P content of alternately grazed and mown grassland were lower than at a surplus of 9 or 18 kg P ha^{-1} yr^{-1} on sand and peat. However, differences between P treatments in DM yield and P content remained constant over the whole period. On the marine clay soil, no differences in DM yield were found between P treatments, but P content in the herbage were lower with balanced P fertilization than with surplus P fertilization. The risk of yield reduction seems to be related to uneven distribution of dung patches and the P buffering capacity of the soil.

Keywords: grassland, grazing, phosphorus, balanced fertilization, dry matter yield, P content

Introduction

In many affluent countries excessive use of phosphorus (P) in the past in agriculture has led to a high P content in the soil, thus threatening the surface water quality by run-off and leaching. In many EU-countries legal limits have been established to protect the water quality. In the Netherlands the P application standard including animal manure and P fertilizer has decreased to balanced P fertilization since 2015. This indicates that P fertilization is limited to the average offtake by crops. Moving towards balanced P fertilization might affect grassland yield and quality, because soil processes might influence the P availability for plant uptake. In the short term, balanced P fertilization is not expected to limit dry matter (DM) production of grass on soils with a sufficient to high soil P (Van der Paauw, 1956). Decreases in P content and P offtake of grass are, however, expected directly after decreasing P fertilization (Power *et al.*, 2005; Schulte and Herlihy, 2007). In the longer term decreases of herbage yield can be foreseen, due to the conversion of available soil P into soil P fractions that are less available to plants.

Grazing is an important factor that influences P flows on grassland. On grazed grassland, herbage P is returned to the soil via excretion of faeces. With balanced P fertilization will have a relatively heavy P supply in manure patches (i.e. a positive P balance), whereas areas without manure patches will have a negative P balance. So far, implications of long-term balanced P fertilization on herbage yield and quality, and on soil P status are not well quantified under grazing conditions. The objective of the long-term field experiment described here, was to examine the effects of balanced P fertilization and two levels of excessive P fertilization, on herbage yield and quality, and on soil P status of grazed grasslands. This paper displays the results of DM yield and P content of grass from 1997 to 2014. The results to 2009 were published on EGF 2012 (Van Middelkoop *et al.*, 2012).

Materials and methods

In the period from 1997 through 2014 an experiment was performed on four permanent grassland sites in the Netherlands: sand (two sites), young marine clay, and peat soil. At the start of the experiment the P soil status was, according to the Dutch standards, for all sites sufficient or high. The experiment had a longitudinal design with six treatments without replicates per site. The treatments were combinations of P and N surpluses, and were randomly assigned to a plot. Fertilization levels were aimed to supply surpluses of 0, 9 and 18 kg P ha^{-1} yr^{-1} (P0, P9 and P18) and 180 and 300 kg N ha^{-1} yr^{-1} (N180 and N300). Cattle slurry and mineral P fertilizer were applied in early spring and before the 4^{th} cut; mineral N fertilizer was applied throughout the whole season. The P9 and P18 surpluses were achieved by applying superphosphate or triple-superphosphate. Superphosphate contains sulphur and was used only in the first five (on peat) or three (on other sites) experimental years. In these years also potassium sulphate was applied on all plots to rule out a sulphur effect. In 2002, the management of the experimental farm on sand_1 was converted to organic farming, implying that P fertilizer was applied as ground phosphate rock (Gafsa-phosphate) which is a less effective P fertilizer than triple superphosphate (Scholefield *et al.*, 1999). Sand_1 ended after 2012, sand_2 ended after 2013. The first and fourth cuts were taken for silage, the other cuts were grazed by heifers or dry cows. DM yield, P content and N content were determined on the day grazing started or cutting took place. The surpluses were calculated as fertilization minus output in silage cuts and in weight increase of livestock. The intake of grass and deposition of urine and excreta during grazing were considered to be an internal cycle.

Differences between treatments in annual DM yields and P and N contents were statistically analysed in a linear model with a fixed and a random part with the Restricted Maximum Likelihood (Reml) method (Harville, 1977), using Genstat (15^{th} edition). The fixed part comprised the experimental treatments as explanatory variables: actual P and N fertilization, number of experimental years, sites, and their interactions. The longitudinal design offered the degrees of freedom to test if the differences were significant. For sand1 the experimental period was split into a conventional period (1997-2001), indicated as sand_1_conv, and an organic period (2002-2012), indicated as sand_1_org.

Results

Mean dry matter (DM) yield responded to P and N fertilization, but responses differed between sites (Figure 1). The response to P fertilization was positive for sand_1_conv, sand_2, and for peat, whereas for sand_1_org and clay no response was found. Effects did not increase or decrease across years. For sand_1_conv, the estimated difference in DM yield compared to 0 kg P surplus ha^{-1} (i.e. balanced fertilization) was 800 kg DM ha^{-1} for P18, which equalled 7% of the estimated DM yield on P0. For sand_2 the difference was 400 kg DM ha^{-1} for P18, which equalled 4% of the estimated DM yield on P0. For peat the difference was 800 kg DM ha^{-1} for P18, which equalled 8% of the estimated DM yield on P0. The estimated response on peat between P0 and P18 was higher than the measured response (Figure 1) because the realized difference in P surplus between P0 and P18 was 15 kg P ha^{-1} instead of the targeted 18 kg P ha^{-1}.

Mean P content responded to P fertilization. P content increased on all sites as a response to P fertilization, except for sand_1_org. On clay the effect of P fertilization was absent in the beginning and positive after two years due to an increase of the response to P fertilization over time. The estimated difference in P content between P0 and P18 was 0.26 g P kg^{-1} DM on sand_1_conv (8%), 0.30 g P kg^{-1} DM on sand_2 (9%), and 0.41 g P kg^{-1} DM on peat (13%). After 18 years the estimated difference on clay was 0.15 g P kg^{-1} DM (6%).

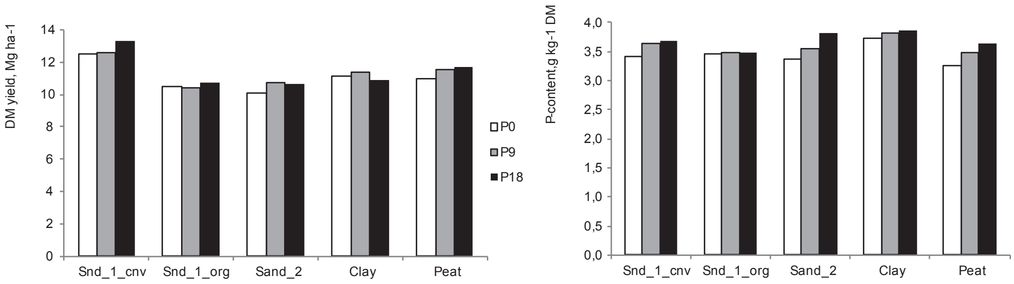

Figure 1. Mean annual dry matter yield (left) and P content (right) at the four sites (sand_1, divided in conventional and organic, sand_2, clay, peat), as function of mean annual phosphorus surpluses in kg P ha^{-1} (P0, P9 and P18), averaged over two levels of nitrogen input, averaged over the period 1997 – 2012 (sand_1), -2013 (sand_2), -2014 (clay and peat).

Conclusions

With balanced P fertilization annual DM yield and P content of alternately grazed and mown grassland were lower than at a surplus of 9 or 18 kg P ha^{-1} yr^{-1} on sand and peat. However, differences between P treatments in DM yield and P content remained constant over the whole period of 18 years. On the marine clay soil, no differences in DM yield were found between P treatments, but P content in the herbage was lower with balanced P fertilization than with surplus P fertilization. These differences between sites seem to be related to the P buffering capacity of the soils. The sand and peat soils had relatively high P-soil capacity parameter and relatively low P-intensity parameter, while the clay soil had relatively a high capacity and intensity parameter. In summary, there is a risk that balanced P fertilization of grazed grassland reduces herbage yield and P content compared to surplus P fertilization, even at relatively high soil P status.

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Changes in herbage nutrient content by dose and type of organic fertilizer

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Abstract

The aim of this study was to assess the effects of organic fertilizers (cow manure + dung water; cattle slurry) applied in different doses of N (54 kg ha^{-1}; 84 kg ha^{-1}; 120 kg ha^{-1}) according to 3 different intensities of grassland utilization (extensive, medium intensive, intensive) on forage quality (nutrients content). The study was performed as a small-plot trial over 5 years in the Czech Rep. Grasslands fertilized with slurry showed significantly higher concentration of crude protein (142.9 g kg^{-1}) than unfertilized (126.4 g kg^{-1}). Extensive grassland utilization significantly decreased the energy value (up to 4.68 MJ kg^{-1} of NEL). Our findings suggest that medium intensive and intensive grassland utilization using organic fertilization which correspond to annual doses of nitrogen of 84 and 120 kg ha^{-1}, are the most suitable for animal nutrition.

Keywords: cattle slurry, fertilization, herbage quality, manure, permanent grassland

Introduction

Organic agriculture relies on ecosystem management and ecological processes rather than on the external flow of agricultural inputs (Foissy *et al.,* 2013). Synthetic inputs are replaced with site-specific management practices to balance input and output nutrients to ensure short-term productivity and long-term sustainability. Hence, organic fertilizers are the irreplaceable foundation for rational agriculture. If applied rationally to grasslands, they can entirely replace mineral fertilizers. In addition, organic fertilizers support soil fertility and have other positive effects (Samuil *et al.,* 2009). Cattle slurry, in particular, is a commonly used fertilizer in many countries, and its effect on grassland has been studied (e.g. Lalor *et al.,* 2012; Duffková and Libichová 2013).

In the Czech Republic, the systematic utilization of organic fertilizers in permanent grasslands is not common, because of their preferred application in intensive arable crops; however, their importance progressively develops mainly in connection with the development of the organic sector. Experimental research is necessary to elucidate this knowledge for Czech local conditions. The objective of this study was to evaluate how the forage quality of permanent grassland is influenced by different applications of organic fertilizers.

Materials and methods

A long-term small plot experiment (one plot size: 12.5 m^2) in completely randomized blocks with four replicates was investigated on permanent grassland in the locality of Rapotín (50°00¢32²N and 17°00¢83²E). The experimental site is situated at 390 m above sea level. Average annual temperature is 7.7 °C and annual precipitation 693 mm. The soil is sandy-loam, *Haplic Cambisol* with horizons Am-Bv-Bv/Cc-Cc. The vegetation of the experimental pasture was classified as *Cynosurion* with some elements of *Arrhenatherion*. Before the experiment setup, the grassland had been used for cattle grazing for over 30 years. Two types of organic fertilizers were applied during 2005-2009: (M) combination of cow manure + dung water and (S) cattle slurry. Organic fertilizers were used in annual doses of nitrogen: 54, 84 and 120 kg ha^{-1}, which approximately corresponded to 0.9 LU.ha^{-1} (LU = livestock unit), 1.4 and 2.0 LU.ha^{-1}. The first 50% dose of the cattle slurry (diluted with water in a ratio 1:3) was applied early in spring and the second 50% after the first cut. Cow manure was applied in autumn, dung water after the first cut. The plots were cut 2-4 times per year depending on the given dose of fertilizer (three intensity levels). Unfertilized plots (F-0) with three intensity levels were also observed as the control treatments: two (extensive), three (medium intensive) and four (intensive) cuts per year. Nutrients in samples (averages of different cuts) (crude protein – CP, crude fibre – CF, ash – A, and ether extract – EE) collected during the vegetation seasons 2005-2009 depending on the term of the cut were analysed according to Czech State Standard 46 7092 (Testing methods of feeding-stuffs). The energy value (ME – metabolizable energy; NEL – net energy of lactation) was predicted by means of the equations officially used in the Czech Republic and the Slovak Republic, which corresponds with the system INRA. Analysis of variance (ANOVA) and LSD test (P<0.05) was used for the statistical data analysis by means of the software Statistica v. 10.

Results and discussion

Regarding the effect of different grassland management on the forage quality (Table 1), we found significant increase of the concentration of CP with increasing intensity of grassland utilization, which

Table 1. Forage quality (content of nutrients in dry matter) of grasslands at different levels of intensity of utilization and fertilization with organic fertilizers in 2005-2009.[1,2]

Traits	**CP**	**CF**	**EE**	**A**	**NFE**	**OMD**	**ME**	**NEL**
	(g kg^{-1})	**(g kg^{-1})**	**(g kg^{-1})**	**(g kg^{-1})**	**(g kg^{-1})**	**(%)**	**(MJ kg^{-1})**	**(MJ kg^{-1})**
Treatments (n=180)								
F-0-ext.	108.9	274.9	27.8	99.8	488.5	61.37	8.34	4.78
F-0-med.int.	120.8	258.1	29.8	111.5	479.8	65.07	8.84	5.13
F-0-int.	139.4	237.9	32.2	110.6	480.0	66.83	9.17	5.36
M-0.9-ext.	112.6	284.1	25.7	96.8	480.9	60.65	8.11	4.62
M-1.4-med.int.	130.4	249.6	31.4	106.3	482.3	65.47	8.81	5.11
M-2.0-int.	154.9	225.9	34.8	112.0	472.3	67.73	9.12	5.32
S-0.9-ext.	116.2	287.1	25.8	99.3	471.6	61.05	8.16	4.66
S-1.4-med.int.	135.2	253.8	31.5	108.2	471.3	65.71	8.83	5.12
S-2.0-int.	162.0	236.2	33.7	120.1	448.0	67.76	9.02	5.26
Standard deviation (S.D.)	18.6	21.8	3.3	7.5	11.6	2.86	0.41	0.28
Means of fertilization types (n=60)								
(F) Nil-fertilization	126.4^{a}	257.0	30.4	107.3	482.8	64.42	8.78	5.09
(M) Cow manure + dung water	137.4^{b}	253.2	31.7	105.0	478.5	64.62	8.68	5.01
(S) Cattle slurry	142.9^{b}	259.0	31.2	109.2	463.6	64.84	8.67	5.01
S.D.	8.4	2.9	0.7	2.1	10.1	0.21	0.06	0.05
Means of intensities of utilization (n=60)								
(ext.) Extensive	112.6^{a}	282.1^{a}	26.4^{a}	98.6	480.3	61.02^{a}	8.21^{a}	4.68^{a}
(med. int.) Medium intensive	128.8^{b}	253.8^{b}	30.9^{b}	108.7	477.8	65.42^{b}	8.83^{b}	5.11^{b}
(int.) Intensive	152.1^{c}	233.3^{c}	33.6^{c}	114.2	466.7	67.44^{b}	9.11^{b}	5.31^{b}
S.D.	19.9	24.5	3.6	7.9	7.2	3.28	0.46	0.32
Factor	***P*-value**							
Year	<0.001	0.283	0.052	0.170	0.359	0.038	0.009	0.009
Fertilization	0.024	0.840	0.874	0.833	0.122	0.952	0.860	0.859
Intensity of utilization	<0.001	<0.001	<0.001	0.089	0.283	<0.001	0.001	0.001
Fertilization*int. of utilization	0.822	0.931	0.702	0.938	0.844	0.992	0.995	0.995

[1] The values in the same column with different superscript letters are significantly different at P<0.05 level for each variable.

[2] Abbreviations: CP, crude protein; CF, crude fibre; EE, ether extract; A, ash; NFE, nitrogen free extract; OMD, organic matter digestibility; ME, metabolizable energy; NEL, net energy of lactation; S.D., standard deviation.

gave on average 152.1 g kg^{-1} DM across years. Fertilization with cattle slurry gave significantly higher herbage concentration of CP (142.9 g kg^{-1} DM) than unfertilized plots (126.4 g kg^{-1} DM). Further, it was found that the extensive grassland utilization gave the highest concentration of CF (up to 282.1 g kg^{-1} DM) and lowest concentration of EE (up to 26.4 g kg^{-1} DM) and energy value (4.68 MJ kg^{-1} DM of NEL). The OMD was negatively influenced by the extensive grassland utilization (61.0, 65.42 and 67.44% for the extensive, medium intensive and intensive grassland utilization, respectively).

These results on forage quality are in agreement with Szewczyk *et al.* (2004), who found that the fertilized (including organic fertilizers) swards produced herbage with 55-60% higher protein concentration than without fertilization. In the experiment of Vintu *et al.* (2008), the influence of organic fertilization was investigated on the content of CP, crude fibre, phosphorus and raw ash. By these authors the applied fertilization systems resulted in an 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.

Conclusions

Our findings suggested that medium intensive (three cuts per year) and intensive grassland utilization (four cuts per year) by the fertilization with doses of organic fertilizers, which corresponded to 84 kg ha^{-1} $year^{-1}$ N and 120 kg ha^{-1} $year^{-1}$ N, respectively, were the most suitable management practices from the viewpoint of animal nutrition. For the specific sites, it is necessary to take into account also the possible environmental risks, which could arise from the application of high doses of organic fertilizers (e.g. decrease of species diversity, leaching of nutrients into ground water). Differences in botanical composition could also influence the results (not presented in this paper).

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Phytoestrogen concentration in red clover (*Trifolium pratense* L.) varieties

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Abstract

Phytoestrogens (PE) are hormone-like plant substances that, when fed, may cause fertility problems, especially noticed in sheep. The aim of the study was to evaluate differences in PE concentration in red clover varieties. Two experiments were performed; Exp 1 with regrowth of 8 red clover varieties harvested at budding and at flowering and Exp 2 with regrowth of 12 varieties harvested at early flowering. There were significant differences between varieties for most PE. In Exp 1, when harvested on the same dates, Peggy, Ilte and SW Torun had higher mean concentration of formononetin, a potent PE, compared to SWÅ RK95097. When considering differences in maturity, Peggy, Ilte and LÖ RK9735 seemed to have a higher content than e.g. SW Ares. In Exp 2, Fregata had high concentration of total PE but the lowest content of formononetin. SW Betty and Ilte had high concentrations of both formononetin and coumestrol. Later harvest (Exp 1) gave decreased concentrations of most of the observed PE. The significant content of coumestrol that was found has not been previously reported for red clover.

Keywords: phytoestrogen, *Trifolium pratense*, variety

Introduction

Phytoestrogens (PE) is a generic term for plant substances that can act as sex hormones in mammals. PE commonly found in red clover are the isoflavones formononetin, daidzein, genistein, biochanin A and prunetin. Coumestrol and lignans are other estrogenic substances that ca be found in plants. Ewes may have different reproductive problems if PE binds to their oestrogen receptors, which eventually can lead to infertility (Adams, 1995). In cattle, the same effect on fertility has not been found (Lundh *et al.*, 1990). However, the content of PE in red clover can be of interest also in milk production. Höjer *et al.* (2012) showed that the content of PE in the milk was relatively high when dairy cows received a diet containing red clover. The concentration of PE varies between different plant parts and developmental stages (Saviranta *et al.*, 2008). Levels are highest in the spring and decreases after flowering (Adams, 1995). The content also can vary between different red clover varieties (Saviranta *et al.*, 2008). The purpose of this project was to examine the differences in PE (isoflavones and coumestrol) concentration in red clover varieties cultivated under Swedish conditions.

Materials and methods

The studies were performed during two seasons. In 2012, eight red clover varieties in pure stand were investigated. In 2014, twelve varieties were sown with the tall fescue/Italian ryegrass hybrid Hykor. All sampling was done in regrowth. Two plots of 0.5 m^2 of each variety were cut. In 2012 sampling was made both in the budding stage (23 July) and in flowering stage (14 August). In 2014 each variety was harvested when it was at the beginning of flowering. Half of each sample was freeze dried and PE were analysed with high-performance liquid chromatography (Steinshamn *et al.*, 2008). The remaining part of each sample was sorted by stage of development and split into leaf sheets, petioles, stems and flowers. The fractions were later merged for nutritional analysis. Data were analysed using SAS mixed model. In addition, the correlation between variables was tested in 2012.

Results and discussion

There were small differences in stage of development between varieties at each harvest time, especially in 2012. Differences in plant parts and nutritional content (NDF, crude fat and digestibility) were small or non-significant.

The detected phytoestrogens are shown in Table 1. In 2012, there were significantly higher concentrations of PE at the first sampling time than in the second. The values in Table 1 are means of the two samplings, except concerning the concentration of coumestrol that was affected by an interaction between sampling time and variety. The reduced concentration of PE at higher stage of development was also correlated to reduced leaf proportion. If taking into consideration the differences in stage of development in 2012, Peggy, LÖ RK9735 and Ilte had higher concentration of formononetin, one of the more potent PE, compared to SWÅ RK95097 and SW Ares. In 2014, there were significant differences in PE concentration between all varieties. Fregata had the highest total concentration due to high content of biochanin A. However, it had the lowest concentration of formononetin and also low concentration of coumestrol

Table 1. Tested red clover varieties and their concentration of phytoestrogens (PE) (g kg^{-1} DM) in 2012 and 2014.[1]

Variety	Sample date[2]	Ploidy[3]	Ley age[4]	Daidzein	Genistein	Formononetin	Prunetin	Biochanin A	Coumestrol		PE total
2012									**23 July**	**14 Aug.**	
SW Ares		2	1	0.07	0.36	6.9^{ab}	0.30^{a}	8.5^{bc}	0.12^{ab}	0.05^{ab}	16.3^{abc}
Peggy		4	1	0.10	0.26	9.4^{b}	0.75^{bcdef}	6.3^{ab}	0.16^{b}	0.05^{ab}	16.9^{abc}
Amanda		4	2	0.10	0.36	7.3^{ab}	0.80^{ef}	7.1^{abc}	0.08^{a}	0.04^{ab}	15.6^{abc}
Ilte		4	2	0.14	0.38	8.9^{b}	0.37^{abcd}	7.5^{abc}	0.16^{b}	0.04^{ab}	17.4^{cb}
LÖ RK9735		4	2	0.10	0.45	8.1^{ab}	0.96^{f}	10.1^{c}	0.14^{ab}	0.04^{ab}	19.7^{c}
SWÅ RK95097		2	2	0.09	0.33	5.3^{a}	0.47^{abcde}	5.0^{ab}	0.10^{ab}	0.03^{a}	11.3^{a}
SW Torun		4	1	0.13	0.37	9.2^{b}	0.36^{abc}	5.8^{ab}	0.13^{ab}	0.06^{b}	15.9^{abc}
SW Torun		4	2	0.12	0.32	8.5^{ab}	0.36^{abc}	5.3^{ab}	0.13^{ab}	0.04^{ab}	14.6^{abc}
SW Yngve		2	2	0.11	0.29	7.3^{ab}	0.59^{abcdef}	3.9^{a}	0.08^{a}	0.04^{ab}	12.3^{ab}
2014											
Atlantis	15 July	4	1	0.06^{ab}	0.48^{abc}	14.8^{de}	0.48^{a}	19.2^{bc}	0.19^{def}		35.3^{bcd}
Callisto	15 July	2	1	0.06^{ab}	0.40^{abc}	11.9^{b}	0.30^{a}	15.2^{ab}	0.15^{b}		28.0^{ab}
Dafila	15 July	2	1	0.07^{ab}	0.52^{bc}	12.6^{bcd}	0.54^{ab}	23.8^{c}	0.16^{bcd}		37.7^{cd}
Fregata	17 July	4	1	0.05^{ab}	0.59^{c}	8.7^{a}	0.76^{abc}	32.7^{d}	0.11^{a}		42.9^{d}
Harmonie	15 July	2	1	0.05^{ab}	0.40^{abc}	12.5^{bcd}	0.33^{a}	15.8^{ab}	0.16^{bcd}		29.3^{abc}
Larus	15 July	4	1	0.08^{b}	0.39^{abc}	14.7^{de}	0.45^{a}	18.6^{bc}	0.18^{cdef}		34.3^{abcd}
Vesna	15 July	4	1	0.07^{ab}	0.34^{ab}	12.4^{bc}	0.40^{a}	14.4^{ab}	0.16^{bcde}		27.8^{ab}
Ilte	23 July	4	1	0.04^{ab}	0.32^{ab}	16.1^{e}	0.56^{abc}	16.6^{abc}	0.20^{f}		33.8^{abcd}
Lasang	23 July	4	1	0.05^{ab}	0.33^{ab}	14.2^{bcde}	1.11^{bc}	12.1^{ab}	0.18^{bcdef}		28.0^{ab}
Lea	21 July	2	1	0.03^{a}	0.27^{a}	11.9^{bc}	0.33^{a}	13.2^{ab}	0.15b^{c}		25.8^{a}
SW Betty	21 July	4	1	0.03^{a}	0.32^{ab}	15.7^{e}	1.26^{c}	9.9^{a}	0.20^{f}		27.4^{ab}
SW Torun	23 July	4	1	0.04^{ab}	0.25^{a}	14.5^{cde}	0.38^{a}	12.1^{ab}	0.19^{ef}		27.5^{ab}

[1] Different lower case letters within each column and year indicate significant differences between varieties.

[2] In 2012 all varieties were sampled on July 23 and August 14. For all PE except coumestrol the presented value is the mean between the two harvests.

[3] 2 = diploid, 4 = tetraploid.

[4] 1 = first harvest year, 2 = second harvest year.

which should result in a relatively low PE effect. The varieties SW Betty and Ilte that are commonly used in Northern Sweden both had high concentrations of coumestrol and formononetin.

The levels of formononetin and biochanin A, and hence the total concentration of PE, were higher in 2014 compared to 2012. It can partly be explained by the relatively big difference in precipitation and temperature between the years. A further difference between the studies was that in 2012 the red clover was grown in pure stand and in 2014 it was mixed with grass. Higher concentrations in mixed crops have also been reported by Kallela *et al.* (1987).

The relatively high levels of coumestrol that was found were unexpected, as in most previous studies of red clover coumestrol has either not been analysed or the concentration has been under the detection limit. We also found a strong correlation between formononetin and coumestrol.

Conclusions

There were relatively large differences between red clover varieties in their content of PE. Because of this, analysis of PE might be introduced in the development and marketing of red clover varieties. Our results also confirm that the development stage is of great importance for the concentration of PE and that there is a variation between years. The significant content of coumestrol that was found has not been previously reported for red clover.

Acknowledgements

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Assessment of forage quality in diverse pastures by sensing spectral reflection and height of swards

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Abstract

Forage quality of grassland swards is a key issue for animal nutrition. Its rapid change in time requires a permanent monitoring to support adequate management decisions. In the present study a field spectrometer (305-1,690 nm) and an ultrasonic distance sensor was applied, able to conduct undisturbed and repeated measurements under field conditions. To evaluate their potential for predicting crude protein (CP) and acid detergent fibre (ADF) content in above-ground biomass, the study was conducted on grassland pasture plots with varying grazing intensities. The potential of two different sensors and the capability of a combination of the sensor data were evaluated against the background of sward structural and functional complexity. Highest prediction accuracies were achieved with hyperspectral calibrations using a partial least square regression method (range of R^2-values: 0.64-0.85 for CP and 0.63-0.75 for ADF). A selection of best fit 2-band normalized difference spectral index (NDSI) from hyperspectral data reduced prediction accuracy, but could be improved by inclusion of ultrasonic sward height. The combination of spectral and distance sensor data mostly achieved or could even exceed calibration accuracies of exclusive hyperspectral data. Thus, sensor fusion allows a feasible option to assess forage quality under field conditions.

Keywords: forage quality, remote sensing, ultrasonic, spectroscopy, sensor combination

Introduction

Pasture quality is highly variable within and between paddocks and during the growing season due to differences in e.g. species composition, sward maturity and soil type (Pullanagari *et al.,* 2012). Conventional techniques of wet chemistry to measure the components of feed quality are expensive and time consuming. In contrast, ground based remote sensing technologies as e.g. spectral canopy reflectance measurements have been recognized as practical means to assess forage quality parameters. Due to costs and complexity of hyperspectral reflectance data, the reduction of the spectral data range by identification of the best spectral bands would facilitate simple sensor applications in the field (Reddersen *et al.,* 2014), but may lead to a loss of prediction accuracy. A combination of spectral data with e.g. sward height measured by an ultrasonic distance sensor (referred to as ultrasonic sward height (USH)) may provide useful information. However, the benefit of such combined sensing technique for a non-destructive determination of forage quality in heterogeneous pastures has yet to be evaluated.

Thus, the goal of this study was to test the performance of spectral reflection data exclusively and in combination with USH for predicting crude protein (CP) and acid detergent fibre (ADF) of pastures, stocked by cows at different stocking densities.

Materials and methods

The research was conducted on a heterogeneous permanent pasture at the experimental farm Relliehausen of the University of Goettingen in the Solling uplands, Central Germany (51°46'55 ' N, 9°42'13 ' E, 180-230 m above mean sea level). The grassland, classified as a moderately species-rich Lolio-Cynosuretum, was continuously stocked by Simmental cows at three stocking densities expressed as standard livestock units (SLU) of 500 kg liveweight ha^{-1} (1) moderate stocking, average of 3.4 SLU ha^{-1}; (2) lenient stocking, average 1.8 SLU ha^{-1}; (3) very lenient stocking, average 1.3 SLU ha^{-1} (Wrage *et al.*, 2012).

In 2013 and 2014 field measurements (sensor measurements and plant sampling) were carried out within one paddock of each stocking density at three samplings (spring, summer and autumn). Subplots (reference plots) (0.25 m^2) were selected in each paddock representing the range of sward compositions and structures (18 samples per paddock and year; duplicate sampling was avoided by GPS measurements). Canopy hyperspectral reflectances were acquired 1 m above surface using a hand-held spectroradiometer (Portable HandySpec Field VIS/NIR, Tec-5 AG, Germany) in the range from 305 to 1,700 nm. USH was assessed with an ultrasonic distance sensor of type UC 2000-30GM-IUR2-V15 (Pepperl and Fuchs, Germany).

Subsequent to sensor measurements, reference plots were clipped at ground surface level (leaving a stubble of about 0.5 cm). Biomass was dried at 65 °C, ground with a 1-mm sieve and measured with a lab near-infrared spectroscopy (NIRS – FOSS XDS). ADF and CP were predicted using specifically developed calibration models at high accuracy levels (ADF: R^2_{cv}=0.97, SE_{cv}=1.45% of DM; CP: R^2_{cv}=0.98, SE_{cv}=0.09% of DM). Field measurements and lab data sets were grouped in a common dataset (n=324) and one subset for each sampling date respectively merged from both years (n=108×3), representing the specific growth characteristics and plant phenology along the vegetation period. Calibration models for ADF and CP were developed either by applying modified partial least squares regression (MPLSR) analysis to the original hyperspectral reflectance using WINISI III (Ver. 1.63, Infrasoft International) or by a selection of best-fit 2-band combinations of the normalized difference spectral index (NDSI) (Inoue *et al.,* 2008) using all possible combinations of two-band reflectance ratios across the available hyperspectral range of 1nm spectral bandwidths. The NDSI was calibrated for exclusive use and in combination with USH. Here, prediction models for ADF and CP were generated by multiple linear regression using the lm-procedure of the R software package (R Development Core Team, 2013).

Results and discussion

USH could only predict ADF in spring with fair accuracy, while CP appeared to be hardly related to sward height. USH-predictions of ADF seem to be strongly linked to growth related stem development in spring, but could barely identify dead material in summer and autumn, which predominantly accumulates in subordinate layers of swards. Both spectral reflection methods gave better results than USH, and could predict CP better than ADF in summer and autumn (Table 1). Hyperspectral data gave better results than the 2-band index NDSI.

Though USH showed a poor prediction accuracy for forage quality, it has a potential to improve predictions of NDSI when used in a sensor combination (Table 1). This is particularly true for ADF, while CP predictions based on solely NDSI already produced very good results, which in summer and autumn could not significantly be improved by USH. Nevertheless, an interaction between both sensor data is evident (Figure 1). Both for ADF and CP, NDSI is the major predictor variable, while USH only plays a role at low levels of NDSI (equivalent to low levels of biomass).

Conclusions

A combination of spectral NDSI and ultrasonic distance sensor (USH) can improve prediction of forage quality compared to their exclusive use and may provide acceptable accuracies for practical application even under heterogeneous pasture conditions. However, methods have to be evaluated with regard to sward development and forage quality parameter.

Table 1. Cross validation (CV) results for predictions of acid detergent fibre (ADF) and crude protein (CP) from exclusive use of ultrasonic sward height (USH), hyperspectral signatures (Hyper) and the normalised difference spectral index with best fit band selection (NDSI) as well as a combination of NDSI and WW2 with USH respectively. Each analysis applied for date-specific swards (N=108 each).[1]

		R^2cv for exlusive use			R^2cv for combined use
		USH	Hyper	NDSI	NDSI+USH
ADF	Spring	0.50***	0.62***	0.45***	0.68***
	Summer	0.29***	0.65***	0.63***	0.66*
	Autumn	0.28***	0.50***	0.46***	0.61***
CP	Spring	0.03*	0.52***	0.41***	0.45*
	Summer	0.03*	0.77***	0.84***	n.s.
	Autumn	n.s.	0.63***	0.60***	n.s.

[1] R^2cv = Coefficient of determination of cross validation; significant at * = 0.05, ** = 0.01, *** = 0.001 probability level. n.s: not significant.

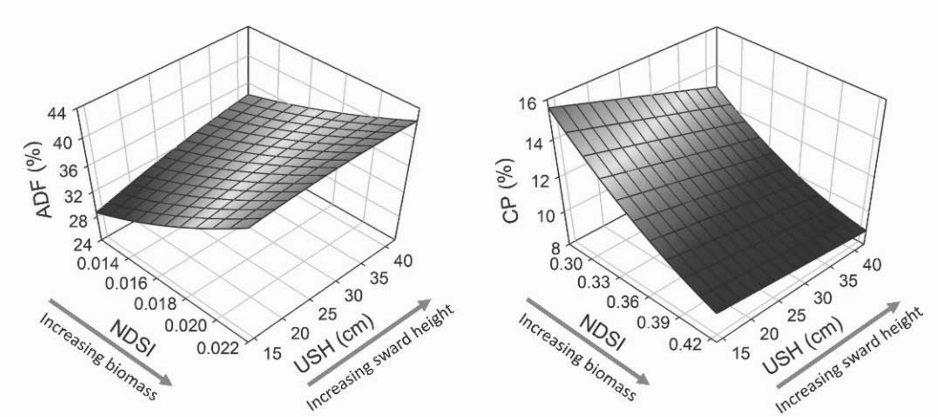

Figure 1. Interactions of sensor combination composed by the 2-band normalized difference spectral index (NDSI) and ultrasonic sward height (USH) on the prediction of acid detergent fibre (ADF) and crude protein (CP) in common swards.

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Silage qualities in the mountain area – a field survey

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Abstract

In mountain areas of Switzerland, economically and environmentally sound dairy production strongly depends on conserved forages of high quality. Due to the lower proportion of high quality grasses and the greater proportion of forbs in mountain grasslands, the ensilability of the forage may be affected. To investigate the silage quality on dairy farms in mountain areas, 31 samples of ensiled forage were collected and analysed for nutrient content and fermentation parameters. On average, the silages had a dry matter (DM) content of 389 g kg^{-1} and contained per kg DM 94 g ash, 130 g crude protein, 303 g acid detergent fibre (ADF), 456 g neutral detergent fibre (NDF) and 5.5 MJ net energy lactation (NEL). They had an average pH of 4.8 with 45 g lactic acid, 9 g acetic acid and also 9 g butyric acid per kg DM. Quality differences of the silages between individual farms were high. The main reasons for reduced quality were the low pre-wilting degree and the high ash content in some samples, as well as a high fibre and a low NEL-content of the forage at ensiling due to progressing sward maturity. The findings indicate the need for an improved farm advisory service.

Keywords: silage, fermentation quality, nutrient contents, mountain areas

Introduction

Because of the shorter vegetation growing period, dairy farming in the mountain area depends much more on conserved forages. To ensure an economically and environmentally sound milk production primarily based on roughage, high quality silage with a well-balanced nutrient content is essential. However, silage production in mountain area is difficult. The lower proportion of high quality grasses, the higher proportion of forbs and the often unstable weather conditions render the production of high quality silage more difficult. Furthermore, difficult terrain conditions may be a source of contamination with soil. The aim of the present study was to survey the silage quality on dairy farms in mountain areas and to identify the main factors which influence the quality.

Materials and methods

The study was carried out on 31 commercial dairy farms in the region Ybrig-Einsiedeln in Switzerland, located between 900 and 1,220 m a.s.l. The silages were produced during summer 2014. Between March and April 2015, samples were taken and nutrient contents and different fermentation parameters were analysed. Seventeen samples were from the first cut and 14 samples from the second and third cut, respectively. The samples were collected from silage stored in bales (15 samples), tower silos (14 samples) or bunker silos (two samples). Only for seven silages had additives been used. Additional data on the silage production practices of the farms was collected with a questionnaire. The quality of the silages was judged together with the farmers using an official Swiss scoring system (Agridea, 2009).

Results and discussion

On average, the silages had a dry matter (DM) content of 389 g kg^{-1}. The variation between the farms was high (Table 1). On average, the silages contained 94 g kg^{-1} DM ash. Four of the samples exceeded the critical value of 110 g kg^{-1} DM. Due to the lower mineral content of the plant material during the

Table 1. Nutrient contents and fermentation parameters of silages from farms in the mountain area.[1]

		All samples				First cut	Second and third cut
		Mean	Standard deviation	Min	Max	Mean	Mean
Number of samples		31				17	14
DM content	g kg^{-1}	389	112	214	641	401	374
Ash	g kg^{-1} DM	94	13	77	138	90	100
Crude protein	g kg^{-1} DM	130	18	99	175	126	134
ADF	g kg^{-1} DM	303	39	247	406	315	289
NDF	g kg^{-1} DM	456	56	377	589	473	435
Sugar	g kg^{-1} DM	80	44	13	171	82	77
NEL	MJ kg^{-1} DM	5.5	0.4	4.3	6.1	5.4	5.6
pH		4.8	0.4	4.1	5.6	4.8	4.8
Lactic acid	g kg^{-1} DM	45	32	3	119	44	47
Acetic acid	g kg^{-1} DM	9	7	1	25	9	9
Butyric acid	g kg^{-1} DM	9	10	0	33	8	11
Ethanol	g kg^{-1} DM	6	3	0	13	6	5
NH_3-N/N total	%	6.1	2.1	2.5	12.3	5.9	6.5
DLG points		70	27	18	100	72	67

[1] ADF = acid detergent fibre; NDF = neutral detergent fibre; NEL = Net Energy Lactation; NH_3-N/N total = ammonia-N proportion.

generative growth cycle, silages prepared from the first cut contained generally lower ash contents (Daccord *et al.*, 2001).

The fibre, crude protein and sugar contents of the silages varied considerably among farms. Samples of the first cut showed higher fibre and lower crude protein contents compared to the second and third cuts. This observation corresponds to the remarks of the farmers in the survey, where many of them stated that the forage of the first cut was ensiled at a rather late maturity stages towards the end of heading and flowering. Consequently, the calculated average NEL content (Net Energy Lactation) of the silages of 5.5 MJ kg^{-1} DM was rather low and varied considerably from 4.3 to 6.1 MJ kg^{-1} DM among farms. Results from the official silage quality analysis 'Raufutterenquête 2014' from all over Switzerland indicated slightly higher average values of 5.7 MJ kg^{-1} DM (Guldimann and Bracher, 2015). The higher NEL content of the silages from the 'Raufutterenquête' may be explained by the fact, that these data predominantly contain samples from the lowlands.

Silages from bales had higher DM contents (430 g kg^{-1}) than silages from tower silos (360 g kg^{-1}). Samples from farms below 1000 m a.s.l. showed higher average DM contents (420 g kg^{-1}) compared to silages from farms above 1000 m a.s.l. (350 g kg^{-1}). However, nutrient and NEL contents were very similar.

The pH ranged between 4.1 and 5.6 (Table 1). In 21 samples, the target pH in relation to the DM content for high quality silage (Nussbaum, 2001) was not reached, most likely due to the low lactic acid production. On average, the silages contained 45 g lactic acid, 9 g acetic acid and 9 g butyric acid per kg DM. A positive correlation between the ash and the butyric acid content was found (r=0.48). This may indicate the importance of contamination with soil.

Classification of the silage quality with the DLG scoring scheme (DLG, 2006) ranged from 18 to 100 points. Out of the 31 analysed samples, 18 samples were classified as good and very good. Six samples had to be classified as defective and seven as bad and very bad. According to the information from the survey, avoiding soil contamination at ensiling was a major challenge. Not only the soil contamination, but also the pre-wiling degree influenced the fermentation quality. Butyric acid was mainly found in silages with low DM contents. A correlation between the DM and butyric acid content was found (r=0.54). A great discrepancy was found between the ranking obtained by the farmers using the official Swiss scoring system and the DLG scoring (Figure 1). None of the farmers classified his own silage as bad or very bad. However, even very good silages were not classified as such by the farmers.

Conclusions

The investigation showed that the quality and the nutrient contents of the silages from mountain areas varied strongly between farms. Harvesting at the optimum maturity stage, avoiding soil contamination and an optimal pre-wiling are the decisive factors for good quality and high nutrient contents. Silage quality can easily be checked with different scoring systems. However, to obtain reliable results experience and practice are needed. The findings indicate the need for an improved farm advisory service.

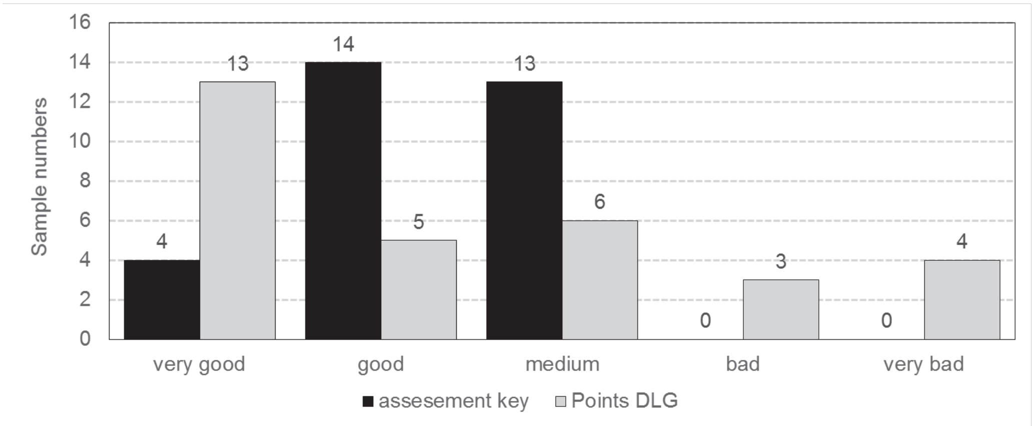

Figure 1. Comparison of the silage quality judged by the farmers with the official Swiss scoring system of Agridea (Agridea, 2009) and the DLG scoring (DLG, 2002).

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Evaluation of fifteen leguminous and non-leguminous forage species to improve forage quality of temporary grasslands in northern Germany

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Abstract

The inclusion of leguminous and non-leguminous herbs in temporary grasslands is a promising strategy to enhance productivity of swards while reducing external inputs. Plants containing phenolic compounds (e.g. tannins) are associated with improved animal health and nutrient use efficiency. However, beneficial properties that promise high nutritive value varied greatly depending on species and environment. The current study aimed to provide a basis of information to identify individual species that beneficially effect ruminant production in northern Germany. Pure stands of eight legumes, six herbs and perennial ryegrass (*Lolium perenne L.*) were sampled in each first full production year 2010 and 2013. Freeze-dried plant material was analysed for crude protein, enzyme soluble organic matter, fibre, total phenolics and condensed tannins. Species variation was evident for each parameter (P<0.001). Apart from sainfoin (*Onobrychis vicifolia L.*) legumes were highest in protein, whereas enzyme soluble organic matter was outstanding in herbs (except plantain (*Planteago lanceolatea L.*)). As expected, phenolic content was generally higher in herbs but concentration of tannins went beyond 1% in sainfoin and birdsfoot trefoil (*Lotus corniculatus L.*) solely.

Keywords: herbs, legumes, temporary grassland, forage quality, condensed tannins, phenolics

Introduction

Previous biodiversity experiments propose that the use of species-rich grasslands is a promising strategy to enhance environmental resource use efficiency without reducing herbage production. Benefits mainly derive from plant community composition and variation in plant functional traits, rather than species richness (Cardinale *et al.*, 2007). However, results under experimental conditions and extrapolation to managed grasslands may be inappropriate. Current research on grass-clover mixtures revealed that swards are advantageous for sustainable intensification of ley farming systems if site-adapted species that diverge in plant functional traits were included (Suter *et al.*, 2015). Although forage quality is the main factor to predict animal performance, recent focus has been on biomass production. The benefits of species-rich alpine pastures for animal performance and product quality derive from dicotyledons evincing high quality (limited in time) and presence of secondary plant metabolites (Jeangros *et al.*, 1999). Forage quality of a broad range of species grown under north German conditions was tested to provide a basis of information for the use in ley systems. It was hypothesized that individual species within groups of leguminous and non-leguminous herbs possess additional features for ruminant production.

Materials and methods

Two independent field experiments including eight legumes, six herbs and perennial ryegrass (Table 1) were established on the research farm 'Lindhof', University Kiel, in northern Germany (54°27'N, 9°57'E). To subject the study to management conditions of organic forage production systems the experimental fields were placed in grass-clover leys within a five-year crop rotation. Species were sown each in pure stands with three replications in a randomized block design. No additional N fertilizer was applied. Each first full production year (2010 and 2013) 200 g of fresh plant material was sampled manually at three harvest times. Unsown species were excluded and samples were immediately frozen.

Content of crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF) and enzyme soluble organic matter (ESOM) were predicted by near infrared reflectance spectroscopy (NIRS) in freeze-dried samples. Total phenolics (TP) were determined by a modulated Folin-Ciocalteau assay based on Singleton and Rossi (1965) using tannin acid as standard. Condensed Tannins (CT) were assayed using a modified butanol-HCL method according to Terrill *et al.* (1992) and expressed as leucocyanidin equivalent. Data of CP, ESOM, NDF and ADF are presented as weighted averages of three harvests. TP and CT data was derived from first harvest samples. Data evaluation was based on a mixed model with year and replication set at random. ANOVA was used to investigate the effect of species and individual species' differences were detected by simple comparisons of mean (Tukey test).

Results and discussion

Investigated forage quality parameters were significantly affected by species ($P<0.001$) and proved broad variation in species ability to converse available resources into high quality biomass.

Most variation were found for CP, ranging from 93 in *Lp* to 230 g kg^{-1} DM in *Th*. Due to nitrogen-fixing legumes, distribution of CP contents according to functional groups was demonstrated (except *Ov*). Findings revealed that *To* may increase CP content of swards in N limited systems if plants were harvested at vegetative stage (following harvests, data not shown). No similar pattern of functional grouping was detected for ESOM and fibre parameters. Apart from *Pl*, ESOM values of herbs were comparable or even exceeded *Lp* and best-performing legumes (*Trifolium*-species and *Mo*). In contrast, ESOM values of *Lc, Ms, Ov* and *Pl* indicated poorer digestibility due to low leaf:stem-ratio and fibrous leaves (mainly in *Pl*) (Jeangros *et al.,* 1999). Although ESOM of most species did not differ from *Lp,* lower NDF contents may increase ruminal fermentation and outflow resulting in higher DM intake (Fulgueira *et al.*, 2007).

For herbs TP contents exceeding 45 g kg^{-1} DM could be demonstrated consistently, whereas most legumes contained less phenolic compounds (Figure 1). CT content of 27 respectively 88 g kg^{-1} DM

Table 1. Annual contents of crude protein (CP), enzyme soluble organic matter (ESOM), neutral detergent fibre (NDF) and acid detergent fibre (ADF) of tested species. Data is derived from a total of 3 harvests and presented as mean values of the two experimental years 2010 and 2013.[1]

Species	Common name	Abbreviation	CP (g kg^{-1} DM)	ESOM (g kg^{-1} DM)	NDF (g kg^{-1} DM)	ADF (g kg^{-1} DM)
Lotus corniculatus L.	Birdsfoot trefoil	*Lc*	219^{abc}	702^{g}	407^{cde}	266^{abce}
Medicago lupulina L.	Black medic	*Ml*	199^{bc}	732^{ef}	381^{ef}	228^{cde}
Melilotus officinalis L.	Yellow sweet clover	*Mo*	205^{abc}	755^{abce}	361^{fghi}	227^{cde}
Medicago sativa L.	Alfalfa	*Ms*	195^{c}	716^{fbg}	407^{bcdef}	229^{abcde}
Oonobrychis viciifolia Scop.	Sainfoin	*Ov*	127^{dh}	719^{f}	385^{ef}	235^{bcde}
Trifolium hybridum L.	Alsike clover	*Th*	230^{a}	754^{bce}	352^{hi}	220^{de}
Trifolium pratense L.	Red clover	*Tp*	199^{bc}	742^{cef}	399^{deg}	243^{abcde}
Trifolium repens L.	White clover	*Tr*	221^{ab}	763^{abc}	376^{f}	231^{bcde}
Achillea millefolium L.	Yarrow	*Am*	127^{deh}	748^{bce}	424^{bcd}	235^{bcde}
Carum Carvi L.	Caraway	*Cc*	115^{def}	787^{ad}	317^{i}	220^{de}
Cichorium intybus L.	Chicory	*Ci*	97^{f}	752^{bce}	375^{feh}	248^{abcde}
Plantago lanceolate L.	Ribwort plantain	*Pl*	110^{ef}	677^{g}	475^{ab}	276^{a}
Sanguisorba minor Scop.	Salad burnet	*Sm*	118^{def}	773^{abd}	426^{bc}	211^{e}
Taraxacum officinale Wig.	Dandelion	*To*	136^{d}	761^{abce}	363^{fehi}	220^{cde}
Lolium perenne L.	Perennial ryegrass	*Lp*	93^{fh}	736^{cdefg}	508^{a}	263^{ab}

[1] Values in the same column with different superscripts differ significantly ($P\leq0.05$).

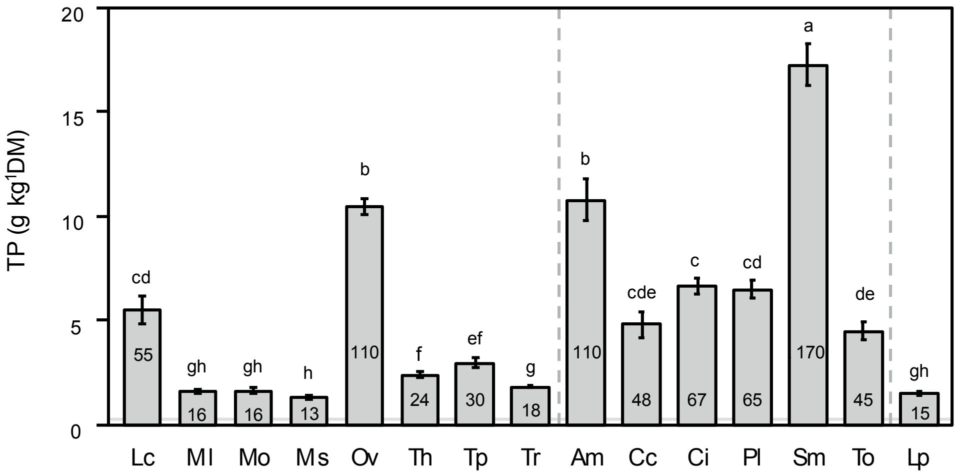

Figure 1. Content of total phenolics (TP) expressed as tannin acid equivalent. Data is derived from first harvest and averaged over both experimental years 2010 and 2013.

may account for exceptional TP in *Lc* and *Ov*. As CT contents exceeding 55 g kg^{-1} may hamper DM digestibility, Ov should not be fed solely. In contrast, CT were significantly lower in *Tp*, *Th*, *Tr*, *Pl* and *SM* (1, 2, 3, 3 and 4 g kg^{-1}) or did not attain more than 1 g kg^{-1} in other species.

Conclusions

Findings provide evidence that individual species differ in plant chemical composition, which might affect forage production under given conditions. As there were some general patterns among functional groups differences between species emphasize that individual quality parameters were not attached to legumes or herbs consistently. Beside the function of nitrogen-fixation, variation of growth type and life cycle accounted for specific difference in species nutritive value.

Acknowledgements

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The effect of previous contrasting grazing intensity on the content of nutrients in pasture forage

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Abstract

The mineral concentration in herbage is one of the key factors that determine its quality. The aims of the study was to determine the optimal period for grazing and the optimal term for hay harvest in relation to the nutrient requirements of cattle and to determine the dynamics of forage yield and mineral concentration during the vegetation season. Besides seasonal development, intensive or extensive grazing treatments applied in the previous grazing seasons significantly affected the mineral concentrations and biomass yields. At the beginning of the vegetation season the concentrations of N, Ca and Mg in the forage were higher in the previously intensively grazed plots than in extensively grazed ones. However, in the course of vegetation season the differences between these two treatments were not significant. Because of low K concentrations and high Ca and Mg concentrations in the forage there was no problem with tetanus ratio (K/(Ca+Mg)). The present work shows that the optimal period for harvest in relation to yield and mineral concentration in the herbage is from mid-May to mid-June in an upland Central European grasslands. After this period changes were revealed in the concentrations of the minerals, which significantly deteriorated the forage quality.

Keywords: grassland, minerals, vegetation season, yield, herbage quality

Introduction

Quality and yield of the forage are the most important factors for the harvest. Generally in the course of the vegetation season the yield is increasing, but the mineral concentration is decreasing (Duru and Ducrocq, 1997). There is a principle question when is the most suitable term to start grazing season or to apply the first cut to fulfil the nutritional and mineral requirements of cattle. Therefore the aim of the study was to determine the optimal period for the grazing and the optimal term for the hay harvest in relation to the nutrient requirements for cattle and determine the dynamics of pasture forage yield and mineral content during the vegetation season.

Materials and methods

The study site is located in the Jizerské hory Mountains in the northern part of the Czech Republic in the village Oldřichov v Hájích. The altitude is 420 m above sea level, the mean annual precipitation is 803 mm, and the mean annual temperature is 7.2 °C (Liberec Meteorological Station). The long-term grazing experiment called the 'Oldřichov Grazing Experiment' established in 1998 was used for the herbage sampling (Pavlů *et al.*, 2007). The dominant species of the sward are: *Agrostis capillaris, Festuca rubra* agg., *Trifolium repens,* and *Taraxacum officinale*. The following treatments were chosen from the long-term grazing experiment: (1) extensive grazing (EG), stocking rate was adjusted to achieve a mean target sward surface height more than 10 cm; (2) intensive grazing (IG), stocking rate was adjusted to achieve a mean target sward surface height less than 5 cm. Both treatments were replicated twice in four paddocks, each of 0.35 ha. The experiment has been continuously stocked by young heifers (initial live weights of 150 to 250 kg) since 1998 from early May until late October. In 2012 the samples of forage biomass were collected weekly from the fenced part of each treatment which was neither grazed nor cut in vegetation season 2012. Six squares of 50×50 cm (different site for each sampling) were cut by electric grass shear in each paddock once a week from 24 April to 3 October (24 weeks of sampling × two treatments × two blocks × six replications are 576 samples in total) dried (48 h at 85 °C) for dry matter (DM) yield assessment and then analysed for N, P, K, Mg, Ca and Na by ICP-OES in an accredited laboratory. Redundancy analyses (RDA) was used for multivariate analyses of mineral concentrations in the herbage, their rations and DM yield. Repeated measures ANOVA was used to evaluate the mineral concentrations in the herbage, their rations and DM yield.

Results and discussion

According to RDA analyses treatment, time and interaction time*treatment explained 3.4% (P<0.001), 17.2% (P<0.001), 20.3% (P<0.001) respectively (P<0.001) of data variability (1st axis) in biomass yield, mineral concentrations and their ratios (Figure 1).

Based on ANOVA (effect of time) concentrations of all studied minerals and K/(Ca+Mg) successively decreased (P<0.001), whilst on the other hand DM yield, Ca/P and K/Na increased (P<0.001) during the vegetation season. In addition, significant treatment effects (P<0.05) were also revealed on yield, the majority of the minerals and their ratios (except for K and ratio K/(Ca+Mg)). It should be noted that according to RDA analyses, the effect of treatment explained five times less variability than the effect of time. Nitrogen is given as an example of this seasonal development (Figure 2) where N concentration in the forage was significantly affected by the treatment (P<0.001), time (P<0.001) and their interaction (P<0.001) (ANOVA).

This progressive decrease of the minerals concentrations simultaneously with increasing of biomass yield during the vegetation season is known as a result of a dilution effect (Duru and Ducrocq, 1997). The optimum mineral concentrations for cattle according to Whitehead (2000) and McDowell and Valle (2000) were found in our experiment in May and June, but the highest yield was found in August and September. A higher proportion of prostrate herbs (*T. repens* and *T. officinale)* at the start of the growing season in IG treatment is a typical response to the long term intensive defoliation management applied in this experiment (Ludvíková *et al.*, 2015) and was probably responsible for the higher concentrations of

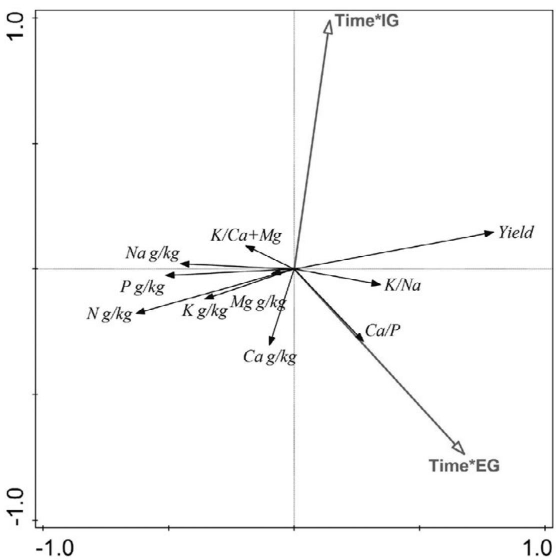

Figure 1. Ordination diagrams showing the result of RDA analysis of mineral concentrations and their ratios in the herbage biomass. * indicate interactions of environmental variables (Time=week of sampling; EG and IG= study treatments).

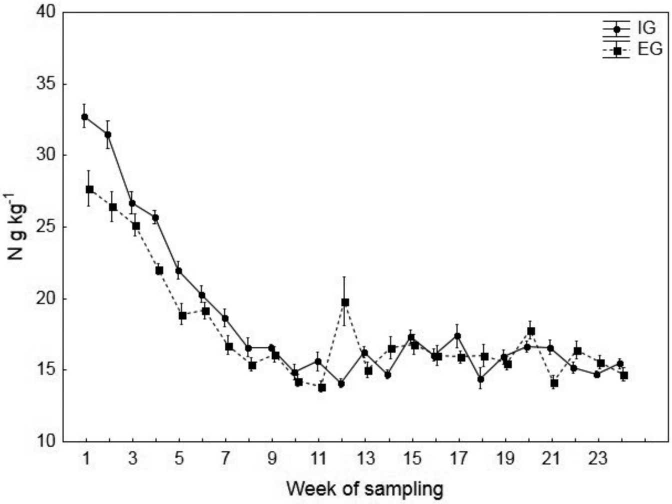

Figure 2. Changes in N concentration in the herbage biomass through the vegetation season in the previously intensively (IG) or extensively (EG) grazed treatment.

Mg and Ca in the herbage. This is because lower concentrations of Mg are usually observed in grasses than in forbs and higher contents of Ca in legumes than in grasses (Whitehead, 2000). Higher concentrations of K (8-15 g kg^{-1} of DM) than the physiological needs for animals were found in both treatments throughout the vegetation season, however the recommended limit 2.5-3.0 of K/(Ca+Mg) (Grunes *et al.*, 1970) was not exceeded.

Conclusions

The effect of previous long term grazing intensity on mineral concentration was revealed at the beginning of grazing season only. The main changes in mineral concentrations in the forage were caused by maturation of sward (dilution effect). The optimum term for grazing or hay cut in relation to yield and mineral concentration in the herbage in an upland grassland is from mid-May to mid-June. After that period, the changes in mineral content significantly deteriorated forage quality. Therefore compensation payment under the environmental schemes should progressively compensate losses in temperate Central European upland grasslands in cases where the first cut is postponed after the middle of June.

Acknowledgements

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The effect of ensiling on variety rank of maize silage

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Abstract

Official variety trials with maize silage determine quality parameters in fresh (i.e. non-preserved) samples. However, farmers use silage as daily component of cattle diet. Quality changes (crude protein and starch content, NDF, organic matter and cell wall digestibility) due to ensiling have been studied. Eight maize silage varieties, frequently grown in Belgium, were monitored at six harvest dates (from 25 to 40% dry matter content) in Merelbeke (Belgium) in 2013 and 2014. At each harvest date, fresh samples were taken and half of the sampled material was ensiled in laboratory silos for 20 weeks. An optimal harvest date was calculated based on frequently measuring starch content and digestibility for both fresh and ensiled samples. All dates not significantly different from the date with maximal values were indicated as optimal. The optimal harvest date(s) did not change after ensiling, but the variety rank based on the silage quality differed from the variety rank based on the fresh quality at this optimal date.

Keywords: maize, silage, variety rank, optimal harvest date

Introduction

The stage of maturity at the time of harvest influences the nutritional quality of ensiled forage maize (Filya, 2004). A harvested produce that is either too wet or too dry makes the silage susceptible to effluent losses and respiration losses, respectively (McDonald *et al.*, 1991). In this respect, harvest date is optimal when the whole-crop dry matter (DM) content is between 30 and 35% (Johnson *et al.*, 1999). Although forage maize is nearly exclusively fed as a silage, reports of official variety trials provide data regarding the fresh (non-ensiled) produce. It is known that quality parameters (including crude protein content, starch content, NDF and organic matter digestibility (OMD)) can fluctuate during ensiling (Lynch *et al.*, 2012). If changes owing to the silage process do not change variety ranks, they are of a minor importance for variety assessments. However as far as we know, very little information is available on this matter. The objective of our study was: (1) to calculate an optimal harvest date for both fresh and ensiled samples of forage maize; and (2) to determine the effects of ensiling on the variety rank at this optimal harvest date.

Materials and methods

Field plots of 8 maize varieties were sown on 24 April 2013 and 17 April 2014 at Merelbeke, Belgium. Plots consisted of four rows with a length of 12 m. Row width was 0.75 m and the plant density was 100,000 plants/ha. The experimental design was a completely randomized block with three replicates. All plots were harvested six times, in a window of DM content between 25 and 40%. Chopped whole-plant samples were subdivided into fresh and ensiled subsamples. Fresh samples were dried at 70 °C for 72 hours to determine DM content and chemical parameters. Ensiled samples were stored in airtight laboratory silos under constant pressure of 200 kg DM m^{-3}. After 20 weeks of storage, ensiled samples were removed from the silos, frozen and transferred into a freeze drier. Compared to oven drying, freeze drying reduces the loss of readily available organic constituents of fermented forages, such as volatile acids. Chemical parameters, including crude protein, starch content, NDF, OMD and cell-wall digestibility were estimated using near-infrared spectra (NIRs) collected at 1,100-2,500 nm at 4-nm intervals using an

Infralyzer 500 spectrophotometer (Bran and Luebbe, Norderstedt, Germany). Calibration equations for the fresh samples were provided by the Walloon Agricultural Research Centre in Gembloux (Belgium). For the ensiled samples, calibration equations were calculated based on chemical analyses of a 10% selection of the samples. Crude protein was determined by the Kjeldahl method. Starch was determined using the procedure of Holm *et al.* (1986). The determination of NDF was based on the laboratory procedures given by Goering and van Soest (1970) using heat-stable amylase and sodium sulfite. Cell wall digestibility, expressed as percentage digestible NDF, is determined after 48h incubation with buffered rumen fluid followed by NDF determination of the undigested residue. The determination of OMD was based on the *in vitro* cellulase technique (De Boever *et al.*, 1997). Optimal harvest date or dates (H_{opt}) was/were calculated for both fresh and ensiled material as the date(s) where starch content and OMD were at maximum. This calculation was done by using a Tukey Test comparing harvest dates with the date showing the highest values for these two parameters. All dates not significantly different from the date with maximal values were indicated as H_{opt}. Quality parameters for fresh and ensiled material at H_{opt} were analysed by analysis of variance (ANOVA) using the statistical program R (version 3.1.1) to indicate significant effects on variety rank. We considered the lack of an ensiling × variety interaction as an indication for a stable variety rank.

Results and discussion

Whole-crop DM content increased linearly during maturation with 2.35% units per week. Ensiling did change the starch content and OMD but the harvest date(s) with maximal values for fresh material also showed maximal values after ensiling. The average DM content for each variety at H_{opt} was between 30 and 35% with an overall mean of 33.5%. This range corresponds with the recommended range suggested by Johnson *et al.* (1999) and was equal for both fresh and ensiled material (Table 1). Therefore, the determination of H_{opt} can be performed by frequently measuring starch content and OMD of fresh material. Because multiple dates were identified as optimal, DM content at H_{opt} varied from 30 to 39% for the individual varieties, which is in accordance with results of Swanckaert *et al.* (2015).

Table 1. Quality parameters of forage maize at optimal harvest date. Effect of ensiling (E), variety (V) and year (Y) on DM content at harvest, crude protein content, starch content, OMD, NDF and cell wall digestibility.

	DM content at harvest	Crude protein content	Starch content	OMD	NDF	Cell wall digestibility
	%	g kg^{-1} DM	g kg^{-1} DM	g kg^{-1} OM	g kg^{-1} DM	g kg^{-1} NDF
Fresh	33.5	69.1	350	747	452	663
Ensiled	33.4	74.3	391	756	321	438
SED[1]	0.42	1.15	7.2	6.2	5.87	21.7
Effect (*P-values)*						
E	0.892	<0.001	<0.001	<0.001	<0.001	<0.001
V	<0.001	<0.001	0.035	<0.001	<0.001	<0.001
Y	<0.001	<0.001	<0.001	0.277	0.344	<0.001
E × V	0.099	0.506	0.499	0.006	0.005	0.149
E × Y	-[2]	0.130	0.704	0.001	–	–
V × Y	<0.001	0.013	<0.001	0.108	0.074	<0.001
E × V × Y	–	0.185	<0.001	–	–	–

[1] S.E.D. = standard error of difference.

[2] The parameter was excluded from the statistical model by stepwise simplification.

We found a stable variety rank for crude protein content and cell wall digestibility. As a consequence, the effect of ensiling for these parameters can be generalized across varieties. Changes due to ensiling were numerically comparable with Lynch *et al.* (2012), who studied six maize types in Ireland on three harvest dates. Compared with fresh material, crude protein content increased in the ensiled produce with 4 g kg^{-1} DM and cell wall digestibility decreased with 225 g kg^{-1} NDF at H_{opt}. We found an unstable variety rank for starch content, OMD and NDF, as evidenced by the significant interaction between ensiling and variety. The silage process increased starch and OMD with respectively 41 g kg^{-1} DM (varying between 15 and 70 g kg^{-1} DM across varieties and years) and 9 g kg^{-1} OM (varying between 2 and 40 g kg^{-1} OM across varieties and years). It decreased NDF with 130 g kg^{-1} DM, varying between 110 and 150 g kg^{-1} DM across varieties and years. As hemicellulose is partially hydrolysed under acidic conditions (Filya, 2004), varieties with a high cell wall fraction tend to lose more hemicellulose than varieties with a small cell wall fraction, leading to a smaller variation between varieties in the ensiled produce. So, although NDF of fresh material differed significantly between varieties, this difference was absent in the ensiled produce.

Conclusions

The optimal harvest date for silage maize can be predicted by frequently measuring starch content and OMD of fresh (i.e. non-preserved) material. Based on the current results, reporting variety ranks without going through the ensiling process continues to be a scientifically justified practice in Belgium for crude protein content and cell wall digestibility. Ensiling changes starch content, OMD and NDF dependent on the variety, resulting in a different variety rank compared to when ranking fresh crops.

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Breeding for better cell wall digestibility in perennial ryegrass (*Lolium perenne*)

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Abstract

The digestibility of forage cell walls is a determinant of animal productivity and efficiency. In our breeding program of perennial ryegrass we aim to improve the cell wall digestibility. In 2010 we have harvested two cuts of 370 non vernalized single plants and we analysed the neutral detergent fibre digestibility (NDFD) and the content of water soluble carbohydrates (WSC). The correlation between the results of the NDFD of the two cuts was 0.48. We selected 63 plants based on their yield, WSC and NDFD content. One polycross with 14 genotypes with a good cell wall digestibility was performed. The NDFD of the 63 plants was reanalysed in 2012 and 2013 on vernalized material. In both years the spring cut and the autumn cut analysed. The correlation between the results of the NDFD of the different cuts was rather low. The progeny of the polycross based on high NDFD was tested in a yield experiment sown in 2012. The NDFD of the progeny was high compared to the NDFD of the standard varieties. Breeding for a better cell wall digestibility seems possible although correlations between the results of the NDFD of single plants analysed in different cuts is low.

Keywords: *Lolium perenne,* cell wall digestibility, quality, breeding

Introduction

Perennial ryegrass is one of the most important forage grasses in Europe because of its high yield potential, good persistence, high protein content and energy value. High levels of water soluble carbohydrates (WSC) can lead to rumen acidosis. Therefor the energy contained within the cell walls, which is released more gradually has become a focus in ryegrass breeding. In our breeding program we aim for a higher quality by improving the cell wall digestibility (NDFD). According to van Parijs *et al.* (2014) it is necessary to differentiate between different organs to be able to improve total plant NDFD. Separating the material in leaf and stem fractions is very laborious. In this study we tried to improve the NDFD without separating the plant material in different fractions. Based on a near infrared spectroscopy (NIRS) analyses, we studied the variation in NDFD of non-vernalized and vernalized plants in pots in several cuts and calculated the correlation between the results. We selected the best plants, allowed them to reproduce and tested the progeny in a field plot experiment.

Materials and methods

In 2010 we grew 370 single non-vernalized plants in pots. The plants were grown from seeds and potted on 1 April. The plants were cut and weighed on 15 June and on 14 July. The samples were dried and we determined the NDFD and the WSC by near infrared spectroscopy (NIRS). The correlation coefficient between the results was calculated.

We selected 63 plants based on their yield, NDFD and WSC content. All selected plants had a total yield of at least the mean of all plants. We selected a group of plants with high WSC and a group with high NDFD. Three polycrosses were composed of plants with high WSC with respectively 13, 22 and 14 components. A fourth polycross was composed of 14 plants with a high NDFD. The progenies of two of the polycrosses, one based on WSC (PCwsc) and one on NDFD (PCndfd), were tested in a yield experiment that was sown on the 5th of April 2012. The control varieties in this experiment were

Abermagic, Cancan, Bovini and Aberavon. The plots were harvested four times both in 2013 and in 2014. We took samples at each cut to determine the NDFD.

The 63 selected plants were replanted in pots in 2012 and the NDFD of these plants was reanalysed in 2012 and 2013 on vernalized material. In both years the spring cut and the autumn cut were harvested and analysed by NIRS. We calculated the mean NDFD of the non-vernalized plants and of four cuts of the vernalized plants and calculated the correlation coefficient between the NDFD values. We calculated the correlation coefficient between the different cuts of the vernalized material.

Results and discussion

The NDFD of the 370 non-vernalized plants varied from 69.4 to 79.9% and from 69.4 to 82.5% in the first and second cut respectively. The correlation coefficient between NDFD values of both cuts was 0.48 and significant (P<0.001).

Table 1 shows the variation in NDFD of the 63 selected plants for the different cuts. The variation was quite high except for the spring cut in 2012 where the NDFD varied from 67.3 to 71.6%. Table 2 shows the correlation coefficient between the NDFD of the 63 selected plants in different cuts and different years. The correlation coefficient between the non-vernalized and the vernalized plants and between the spring and autumn cuts was low: 0.19 (not significant) and 0.26 (sign P<0.05) respectively. The

Table 1. Variation in digestibility of the neutral detergent fibre (NDFD) of 63 plants in different cuts on non-vernalized (2010) and vernalized (2012/2013) material.

	Cut	NDFD mean (%)	NDFD coef. var. (%)
Non vernalized plants	2010 cut 1	74.1	2.70
	2010 cut 2	74.7	4.41
Vernalized plants	2012 spring	69.9	1.29
	2012 autumn	66.7	2.84
	2013 spring	76.9	2.55
	2013 autumn	72.6	3.14

Table 2. Correlations between NDFD values of 63 plants analysed in several cuts in 2010 (non-vernalized) and in 2012 and 2013 (vernalized) (significant at P<0.05 in bold).

		2010	Spring cut			Autumn cut			Total		
			2012	2013	12-13	2012	2013	12-13	2012	2013	12-13
2010		1.00	0.44	0.02	0.18	0.18	0.02	0.13	0.35	0.03	0.19
Spring	2012	0.44	1.00	0.38	0.67	0.02	-0.03	0.02	0.44	0.19	0.37
	2013	0.02	0.38	1.00	0.94	0.18	0.30	0.31	0.32	0.77	0.72
	12-13	0.18	0.67	0.94	1.00	0.15	0.23	0.26	0.42	0.69	0.71
Autumn	2012	0.18	0.02	0.18	0.15	1.00	0.38	0.79	0.90	0.37	0.66
	2013	0.02	-0.03	0.30	0.23	0.38	1.00	0.86	0.34	0.84	0.74
	12-13	0.13	0.02	0.31	0.26	0.79	0.86	1.00	0.72	0.75	0.86
Total	2012	0.35	0.44	0.32	0.42	0.90	0.34	0.72	1.00	0.41	0.75
	2013	0.03	0.19	0.77	0.69	0.37	0.84	0.75	0.41	1.00	0.91
	12-13	0.19	0.37	0.72	0.71	0.66	0.74	0.86	0.75	0.91	1.00

correlation coefficient between the results of 2012 and 2013 was 0.38 (sign. P<0.01), 0.38 (sign. P<0.01) and 0.41 (sign. P<0.001) for the spring cuts, the autumn cuts and the average of all cuts, respectively.

Figure 1 shows the results of the NDFD of the progenies of PCwsc and PCndfd tested in the yield experiment compared to the standard varieties in 2012 and 2013. The average NDFD over the two years of the best standard variety Abermagic is 69.2% compared to 71.2% for PCndfd and 69.4% for PCwsc. The progenies of PCndfd had the highest NDFD in all cuts except the third cut in 2013. In this cut the variation was very low (67.1-68.5%).

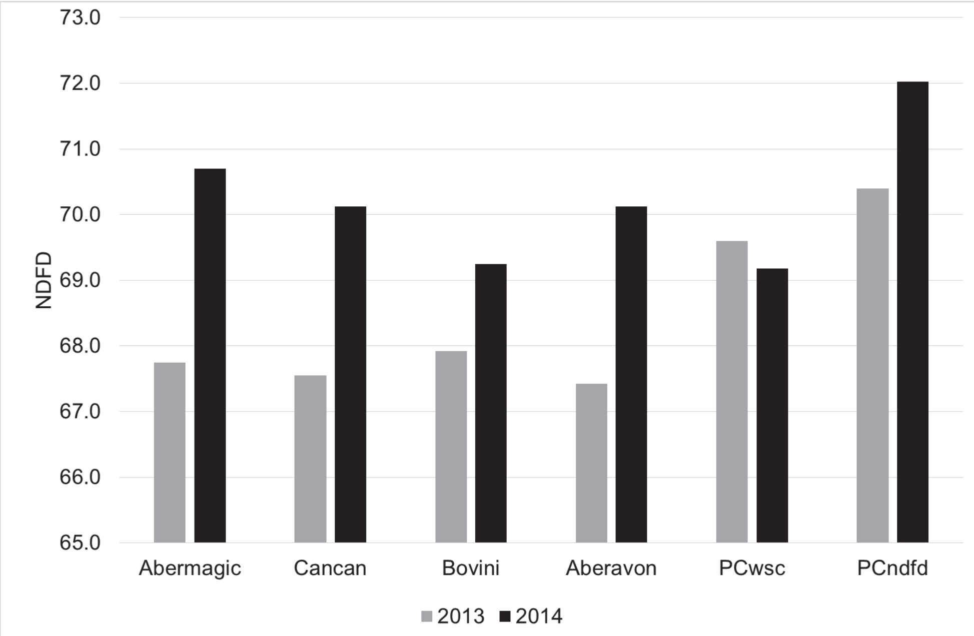

Figure 1. Results of the NDFD (%) in 2013 and 2014 of the progenies of two polycrosses (PCwsc – high WSC and PCndfd – high NDFD) compared to standard varieties Abermagic, Cancan, Bovini and Aberavon. Data are averages over 4 cuts taken in 2013 and 2014.

Conclusions

One of the targets of the perennial ryegrass breeding programme of ILVO is the development of varieties with improved cell wall digestibility. The variation in NDFD is quite high but the correlations between different cuts and years, although positive, are rather low. Non-vernalized plants that were selected for a high cell wall digestibility had a progeny that showed a higher NDFD compared to a set of commercialized varieties. Breeding for a better cell wall digestibility therefore seems possible. It seems unnecessary to separate the plant material in different fractions to select plants with a higher NDFD.

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Influence of early cutting dates on forage yield and quality of alpine pastures

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Abstract

Alpine pastures are characterised by a diverse botanical composition managed at graduated intensity. Rapid grass growth at the beginning of the vegetation period generally results in fast decline in forage quality. The reduced net energy lactation (NEL) content of the forage complicates its utilisation for higher yielding dairy cattle. The aim of our study was to investigate the influence of early utilisation dates on forage yield and the quality of alpine pastures managed at different intensities. Pastures established for a 60-year-long fertiliser experiment located at 1,400 m a.s.l. were used. The first cut of selected plots (zero fertiliser, NPK and PK) was cut standard (usual beginning of summer pasture), early (21 days before usual beginning of summer pasture) and very early (35 days before usual beginning of summer pasture). The highest dry matter yields (DMY) were found for the standard cutting treatment. At the early and very early cutting dates (21 resp. 35 days before usual beginning of summer pasture) the DMY was significantly influenced by the fertiliser treatment. A significant interaction between fertiliser and cutting treatment was found. DMY was highest for NPK swards and lowest for 0 swards. NEL content was highest at the first cut for the early and very early cutting regimes. The results demonstrate the potential to improve forage quality by earlier utilisation, but also indicate consequences for yield from pastures managed at higher intensity.

Keywords: alpine pasture, fertilisation, cutting date, productivity, nutritional value

Introduction

Thanks to the forage produced on Swiss alpine pastures, an additional 10% of livestock can be fed (Herzog *et al.*, 2013). As a result of the well-managed grazing activities, alpine pastures provide a broad range of ecosystem services. However, during the past decades grassland utilisation in alpine areas has declined and shifted towards more extensive utilisation, leading in some areas to serious scrub and tree encroachment (Mack, 2008). The decline in milk yield during the summer alpine pasture period, with its consequent economic impact, is one of the most important factors in decreasing grassland utilisation in alpine areas (Fischer, 2011). Rapid grass growth at the beginning of the vegetation period results in fast decline in forage quality (Brühlmann and Thomet, 1991) with strongly reduced net energy lactation (NEL) content. The usual beginning of summer pasture on alpine pastures is generally rather late due to risk of a late winter onset and traditional habits. If utilisation of alpine pastures by dairy cows is to be maintained in the future, solutions to improve the inadequate energy supply at the beginning of the summer pasture have to be developed. In this study we examined the influence of different utilisation dates on forage quantity and quality of alpine pastures differing in botanical composition and fertiliser supply.

Materials and methods

The study was carried out at a 60-year-long fertiliser experimental site located on the alpine pasture of the Eggenalp (Zweisimmen BE, Switzerland, 46°33'54' N 7°21'27'E) 1,340 m a.s.l. with an acidic brown earth soil. The original fertiliser experiment involves eight different fertiliser treatments on 5×10 m plots with swards of different botanical compositions in a randomised complete block design with four replications. The three selected treatments (0, NPK, PK) simulated different levels of management intensity. The various fertilisation treatments received the following amount of nutrients: 82 kg N ha^{-1}

and 180 kg K_2O ha^{-1} each year and 180 kg P_2O_5 ha^{-1} each third year. This lead to subsequent differences in the abundance of grasses, legumes and forbs for the different treatments 0: 43/15/42%, NPK: 55/9/36%, PK: 48/21/31% (assessed with the frequency method of Daget and Poissonet (1969)). Each selected plot was split into subplots where three cutting regimes (very early, early, standard) were applied. In this way, various utilisation dates of first and subsequent utilisations (sample dates cf. Table 1) with respect to the common start date of the summer pasture period (very early: -35 d before, early: -21 d before, standard: 0 d) are simulated. The annual rainfall at the experimental site was approximately 1,900 mm with an average annual temperature of 4 °C. Sward biomass was determined by mowing a 0.7×9 m subplot at a height of 4 cm at each sample date of each fertiliser replication. Dry matter yield (DMY) was determined on a subsample of 300 g by drying for 24 hours at 105 °C. Samples for nutritional analysis were dried for 24 hours at 55 °C before processing. Nutritional content (acid detergent fibre (ADF) and crude protein (CP)) was determined by NIRS and additionally verified by wet chemistry analysis (Agroscope, 2015). NEL was estimated by regression equations in accordance with Daccord *et al.* (2013). Statistical analysis using the statistical software R (R Development Core Team, 2010) was carried out for each sample date separately by fitting a linear mixed model with a random intercept for the block factor and fixed effects for fertiliser, cutting treatment and their interaction.

Results and discussion

The NPK treatment resulted in the highest and the 0 treatment in the lowest total DMY (Table 1). A significant effect of the fertiliser treatment on DMY was found at sample date one, two, and four. At date three a significant interaction between fertiliser and cutting treatment could be observed. This indicates the dissimilar response of the different swards to the different cutting regimes for the first cut and highlights the importance of the time of the first cut for the total DMY. For the fertilised plots (NPK, PK), the very early and early cutting treatment was similar and resulted in a notably lower total DMY than the standard treatment. The NPK sward was dominated by early and productive grasses such as

Table 1. Net energy for lactation (NEL), acid detergent fibre (ADF), crude protein content (CP) and dry matter yield (DMY) of different fertilised (F) swards of a 60-year-old fertiliser experiment on an alpine pasture managed under a standard (first utilisation at usual beginning of summer pasture), an early and a very early (first utilisation 21 resp. 35 days earlier) cutting regime (C).[1,2]

Treatment		Sample date 1: -35 days				Sample date 2: -21 days				Sample date 3: 0 days/standard				Sample date 4: +35 days				Total
C	F	NEL[1]	ADF[2]	CP[2]	DMY[3]	NEL[1]	ADF[2]	CP[2]	DMY[3]	NEL[1]	ADF[2]	CP[2]	DMY[3]	NEL[1]	ADF[2]	CP[2]	DMY[3]	DMY[3]
Very early	0	6.25	234.8	159.1	0.4	6.26	226.0	151.6	0.6	5.75	288.2	150.7	4.0	5.724	276.8	163.6	6.0	10.4
	NPK	6.44	200.4	207.9	5.1	6.20	201.3	189.1	1.9	5.70	300.7	170.2	10.2	6.073	259.4	202.9	14.4	31.6
	PK	6.30	192.4	185.3	1.2	6.12	208.9	175.8	0.7	5.69	299.6	169.5	8.5	5.863	267.4	167.7	10.5	20.5
Early	0					5.90	226.8	148.5	2.9	5.63	299.2	149.2	5.8	5.772	271.9	160.6	5.7	14.4
	NPK					6.14	214.7	170.6	8.7	5.74	296.8	157.9	8.3	5.898	268.2	192.8	13.6	30.6
	PK					6.02	216.4	159.1	3.5	5.75	297.9	164.6	9.0	5.848	271.0	171.7	8.9	21.4
Stan-dard	0									5.35	307.4	133.3	9.4	5.645	279.8	158.5	6.6	16.1
	NPK									4.98	340.0	114.3	34.5	5.883	262.4	187.4	15.5	50.0
	PK									5.32	317.1	135.9	18.2	6.091	247.3	180.8	9.0	27.2
P-value	C	–	–	–	–	0.303	0.250	**	***	***	**	**	***	0.805	0.450	0.642	0.528	***
	F	0.367	**	***	***	0.305	0.230	***	*	0.410	0.259	**	***	**	*	***	***	***
	CxF	–	–	–	–	0.883	0.810	0.426	0.126	0.236	0.516	**	***	0.125	0.199	*	0.806	**

[1] NEL: MJ NEL kg^{-1} DM; ADF: g kg^{-1} DM; DMY: kg DM ha^{-1}.

[2] Sign. *** P<0.001, **P<0.01, *P<0.05.

Dactylis glomerata L. and *Festuca pratensis* Huds.. PK and 0 swards were characterised by fewer grasses and grasses with later heading dates such as *Festuca rubra* L. or *Agrostis capillaris* L. and a higher proportion of legumes. The high NEL content of 6.2-6.4 MJ kg^{-1} DM for all fertiliser treatments at sample date one show the potential of considerable milk yields from the forage for the very early cutting treatment. In contrast, at sample date three the NEL content of the standard treatment was substantially lower (NPK: 5.0 MJ NEL kg^{-1} DM, PK: 5.3 MJ NEL kg^{-1} DM and 0: 5.4 MJ NEL kg^{-1} DM). The significant higher ADF content of the NPK swards for the standard cutting regime resulted in a pronounced decline in the NEL content. This was most likely the effect of an increased lignification of the early grasses intensified through N fertilisation (Caputa, 1966). Similarly the slower drop in CP due to a higher proportion of legumes and forbs led to a distinctly slower decline in the NEL content for PK and 0 treatments (Schubiger *et al.,* 1998). At date four there was a significant effect of fertiliser treatment for NEL, ADF and DMY, but no effect of cutting treatment, indicating the decreasing importance of the cutting regime when the preceding cutting date was the same.

Conclusions

Regarding the use of alpine pasture, the importance of the date of the first cut depends on the characteristics of the sward. While intensive swards (high nutritional status, high proportion of early grasses) require earlier utilisation to provide forage of a high quality, less intensive swards are more flexible with respect to yield and quality changes at differing first utilisation dates. The results demonstrate the potential to improve forage quality by earlier utilisation, but also indicate the lower yield as a consequence for pastures managed at higher intensity.

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Dry matter intake and in vivo digestibility of different cereal-legume intercrops mixtures in sheep

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Abstract

There is renewed interest in using cereal-legume mixtures silages in ruminant ration to assist forage self-sufficiency and climate change adaptation. However, data on feed value of cereal-legume silages remains limited in Europe. The objective of this study was to evaluate dry matter intake (DMI) and organic matter digestibility (OMD) of three cereal-legume silages harvested at two stages of growth using Texel sheep. The three mixtures were wheat with pea and vetch (WPV), bearded triticale with pea and vetch (+TPV) and beardless triticale with pea and vetch (-TPV). The DMI was on average 65.4 g kg^{-1} $BW^{0.75}$ for silages harvested at early stage without significant difference between mixtures; the DMI was lower for silages harvested at late stage (28.0, 37.5 and 46.2 g kg^{-1} $BW^{0.75}$ for +TPV, –TPV and WPV respectively). The OMD for WPV and +TPV was higher than for –TPV at both growth stages. Finally, the digestible organic matter intake was on average 41.5 g kg^{-1} $BW^{0.75}$ at early stage, but was lower for +TPV and –TPV (16.8 and 21.1 g kg^{-1} $BW^{0.75}$) than for WPV (28.1 g kg^{-1} $BW^{0.75}$) at late stage. Overall, these results suggest that the cereal-legume silages harvested at early stage have a higher nutritive quality.

Keywords: *in vivo* digestibility, dry matter intake, cereal-legume silages, sheep

Introduction

Intercropping is an old practice used in many areas of the world. This practice, especially those employing cereals with legumes, is developing in French ruminant production systems to permit forage self-sufficiency, reduce N fertilizers and for climate change adaptation. Several environmental, agronomic and economic interests of cereal-legume intercrops mixtures have been reported (Pelzer *et al.*, 2012). Combining the growth of cereal forages with legumes enhance also the forage yields and crude protein (CP) concentrations in comparison with cereals whole crops or grass silages (Adesogan *et al.*, 2002). Previous works have compared few cereal-legume silages in terms of nutritive values and fermentation characteristics (Adesogan *et al.*, 2002; Mustapha and Seguin, 2004). These works have shown that fermentation pattern and nutritive value were first affected by stage of growth at harvest and the proportion of legume to cereal. Despite these works, data on feed value of cereal-legume silages remain limited in Europe, mainly due to a lack of *in vivo* measurements and the potential vast number of mixtures used. Thus, the objective of this study was to evaluate the effect of three common cereal-legume silages used in France and harvested at two stages of growth on dry matter intake (DMI) and organic matter digestibility (OMD).

Materials and methods

Three 5-ha plots were sown at the INRA experimental farm of Lusignan (France) with associations of field pea (*Pisum sativum* L., cv. Assas), common vetch (*Vicia sativa* L., cv. Savane) and cereal at the seed ratio of respectively 17, 20 and 220 seeds m^{-2}. The cereals used were wheat (*Triticum aestivum* L., cv. Soisson), bearded triticale (*X Triticosecale*, cv. Ragtac) or beardless triticale (*X Triticosecale*, Agri Obtentions). The three mixtures were harvested at two stages: early (20 May 2010, cereal at the end of stem elongation) and late (22 June 2010, cereal at dough-milky stage). Crops harvested at early stage were wilted before ensiling to target 25-30% dry matter (DM) content whereas at late stage, crops were directly ensiled in experimental silos of approximately 450 kg DM and then stored before *in vivo* evaluation. Just before harvesting, the botanical composition of each mixture was determined from three randomly chosen 1 m^2 plots.

The *in vivo* study was carried out at the INRA experimental farm of Le-Pin-au-Haras (France). Six Texel male castrated sheep (on average 61.3 kg at the beginning of the study) were used to determine the OMD and DMI of the six silages. Two measurement periods per forage type were carried out successively. After an adaptation period of 14 days, a 5-days digestion trial was performed followed by 7 days off and then a second 5-days digestion trial. For each digestion trial, individual daily DMI was measured and total faeces collected and weighed. Silages were fed *ad libitum* in two meals. Offered forage and refusals were recorded daily and representative samples of the silage offered and refused, and of faeces from each sheep were collected daily. These samples were pooled at the end of the trial into: one sample of offered silage per forage type, one sample of refusal and one sample of faeces per sheep and forage. Faeces and refusal samples were analysed for ash, whereas silages were analysed for ash, CP (Rapid N Cube, Elementar) and for neutral detergent fibre (NDF) and acid detergent fibre (ADF) content using an Ankom fibre analyser (Ankom Technology) with heat-stable amylase. Data were analysed using the mixed procedure of SAS. Data for chemical composition were analysed with 'silage', 'growth stage' and 'silage × growth stage' as fixed effects. Data for *in vivo* measurements were analysed with 'silage', 'growth stage' and 'silage × growth stage' as fixed effects and 'sheep' as random effect. Multiple comparisons of means were performed with an adjusted Tukey-Kramer test.

Results and discussion

Despite the use of the same seed ratio for the three crops, the legume proportion was greater for the silages with wheat (WPV) than for silages with triticale: 0.32 and 0.41 for WPV at early and late stage, respectively vs 0.14 and 0.19 at early stage and 0.14 and 0.27 at late stage for the silages with triticale without and with spikes. Consequently, the CP content for WPV silages was higher than CP contents for silages with triticale at both stages (Table 1). For silages with triticale, the CP content decreased with maturity at harvest whereas for WPV the CP content at early stage (132 g kg^{-1} DM) was lower than that at late stage (142 g kg^{-1} DM). The higher legume proportion at this growth stage can partially explain these values. The NDF and ADF contents were similar between silages and stages of growth (P>0.10).

The DMI and OMD results are shown in Table 2. The DMI was on average 65.4 g kg^{-1} $BW^{0.75}$ for the three silages harvested at early stage. The DMI decreased with maturity at harvest for all mixtures (P<0.001) and especially for the bearded triticale+pea+vetch silage (+TPV): 28.0 g kg^{-1} $BW^{0.75}$ vs 46.2 and 37.5 g kg^{-1} $BW^{0.75}$ for WPV and the beardless triticale+pea+vetch silage (-TPV) respectively. These intake values were lower than DMI measured with pea+wheat silages at late stage (on average, 51 g kg^{-1} $BW^{0.75}$, Salawu *et al.,* 2002). The low intake level for +TPV at late stage, was probably due to palatability problems with the triticale related to the presence of barbs (Scerra *et al.,* 1994). The OMD was higher

Table 1. Dry matter (DM, g kg^{-1}) and chemical composition expressed as g kg^{-1} DM of the three cereal-legume silages (+TPV: bearbed triticale+pea+vetch; -TPV: beardless triticale+pea+vetch and WPV: wheat+pea+vetch) harvested at two stages of growth.[1]

	Early stage			Late stage				Effects		
	+TPV	-TPV	WPV	+TPV	-TPV	WPV	SEM	Sil	Gs	Sil×Gs
DM	207[b]	269[ab]	268[ab]	240[ab]	277[a]	241[ab]	11.0	0.011	0.60	0.08
CP[a]	120[ab]	102[bc]	132[a]	98[bc]	84[c]	142[a]	5.0	<0.001	0.05	0.04
NDF[a]	561	548	525	550	498	510	24.1	0.33	0.23	0.72
ADF[a]	352	317	317	332	302	306	16.5	0.27	0.20	0.80

[1] CP = crude protein; NDF = neutral detergent fibre; ADF = acid detergent fibre; Sil = effects of the types of silage; Gs = effect of the growth stage at harvest; SEM = standard error of means.

Table 2. Dry matter intake (DMI), *in vivo* organic matter digestibility (OMD) and digestible organic matter intake (DOMI) of the three cereal-legumes silages (+TPV: bearbed triticale+pea+vetch; -TPV: bearbless triticale+pea+vetch and WPV: wheat+pea+vetch) harvested at two stages of growth.[1]

	Early stage			Late stage				Effects		
	+TPV	-TPV	WPV	+TPV	-TPV	WPV	SEM	Sil	Gs	Sil×Gs
DMI, kg d^{-1}	1.44^a	1.42^a	1.47^a	0.55^c	0.82^b	0.91^b	0.08	<0.001	<0.001	<0.001
DMI, g kg^{-1} $BW^{0.75}$	65.2^a	65.4^a	65.5^a	28.0^d	37.5^b	46.2^c	2.85	<0.001	<0.001	<0.001
OMD	0.69^a	0.67^b	0.71^a	0.64^b	0.59^c	0.65^b	0.008	<0.001	<0.001	0.20
DOMI, g kg^{-1} $BW^{0.75}$	41.1^a	40.5^a	42.9^a	16.8^c	21.1^c	28.1^b	1.69	<0.001	<0.001	<0.001

[1] Sil = effects of the types of silage; Gs = effect of the growth stage at harvest; SEM = standard error of means

for WPV and +TPV than for –TPV whatever the stage of growth (P<0.001). Otherwise, for all silages, the OMD decreased with the maturity at harvest (P<0.001) and varied from 0.71 to 0.59. These OMD values were lower than OMD found in Feed Table for grass silages (INRA, 2007), but were consistent with OMD previously measured on cereal-legume silages (Salawu *et al.*, 2002; Arrigo *et al.*, 2014 and 2015). The DOMI was on average 41.5 g kg^{-1} $BW^{0.75}$ at early stage, but was lower for +TPV (16.8 g kg^{-1} $BW^{0.75}$) and –TPV (21.1 g kg^{-1} $BW^{0.75}$) than for WPV (28.1 g kg^{-1} $BW^{0.75}$) at late stage. The very low DOMI values for silages harvested at late stage suggest that these silages, especially those with triticale, had a poor nutritive quality.

Conclusions

The results of this study show that for early harvesting silages, the feed value of the three cereal-legume mixtures was similar. At late growth stage, the feed value of the mixture with wheat was higher than the feed value of mixtures containing triticale. Overall, the results suggest that silages harvested at early stage have a higher nutritive quality than silages harvested at late stage. However, forage yields are generally lower at early stage compared to late stage.

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Wavy hair-grass (*Avenella flexuosa*) – yield, regrowth and feed quality

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Abstract

Wavy hair-grass *(Avenella flexuosa* (L.) Drejer; *Deschampsia flexuosa* (L.) Trin.*)* is the main pasture species in blueberry mountain birch forest and dwarf birch – blueberry moorland, which cover large parts of outfield pastures in the mountainous region of Southern Norway. Blueberry mountain birch forest with continuous mats of *A. flexuosa* was fenced in and harvested at different times in the summers of 2014 and 2015. Regrowth was also recorded. The grass from sample plots was dried after harvest, and analyzed for feed quality using NIRS. There were no statistically significant differences in total net energy yield between different harvesting regimes. Grass growth was highest in early summer, and harvesting on 2 July gave about 60% of the seasonal yield. Grass yield in undisturbed population increased until the last harvest (early September). Regrowth after harvest was small at the end of the season, but the growth here corresponded with the growth in undisturbed population. *A. flexuosa* remained at vegetative stage during the season. The energy value was highest at harvest first in July, and relatively constant at later harvests. The protein concentration declined towards the end of the season.

Keywords: *Deschampsia flexuosa*, growth rate, rough grazing land, boreal forest

Introduction

Wavy hair-grass *(Avenella flexuosa* (L.) Drejer; *Deschampsia flexuosa* (L.) Trin.*)* is a perennial, tussock-forming grass with a branched system of rhizomes forming lateral shoots and adventitious roots. It thrives and flowers abundantly in full light, but can endure very low light intensities; under canopy mat sparser, patchy and without flowering shoots (Scurfield, 1954). The way of vegetative reproduction can cause it to form dense, continuous mats in some vegetation types, such as blueberry forest. It is the main pasture species in blueberry mountain birch forest and dwarf birch-blueberry moorland, which cover large parts of outfield pastures in the mountainous region of Southern Norway. Yield, quantity and quality of *A. flexuosa* mats, and how this is affected by defoliation (simulated grazing) in the grazing season, is of great importance for grazing capacity in these areas.

Materials and methods

A study was carried out in blueberry mountain birch forest with high coverage (continuous mats) of *A. flexuosa*, about 860 m a.s.l. in Vingelen, Hedmark County, Norway (62° 31' N, 10° 47' E). The experimental area is normally covered in snow until mid-May. The grazing period is usually from mid-June to early September. At the nearest situated meteorological station – Røros Lufthavn, 652 m a.s.l. 30 km northeast of Vingelen – normal mean temperature is 5.6 °C in May, 10.1 °C in June, 11.4 °C in July, 10.4 °C in August and 6.1 °C in September. The experiment was established in a randomized complete block design with four replicates, duplicates within each replicate and plot size of 0.12 m^2. Plots within each replicate were put out side by side, and replicates were placed within an area of 0.2 ha. Four harvest regimes were studied: (A) harvesting 2 July and 2 September; (B) harvesting 2 August and 2 September; (C) harvesting 2 July, 2 August and 2 September; (D) harvesting 2 September. The experiment was conducted with two one-year fields, one in 2014 and one in 2015. Plant material above a stubble height of 1 cm was harvested using scissors and some hand picking. Dead plant material and any other species than *A. flexuosa* were removed before and under harvesting. Harvested plant material was dried at 60 °C, weighed, and feed quality was determined by NIRS analysis at NIBIO Løken where a broad calibration for grassland (n>1,500) was applied (Fystro and Lunnan, 2006). Most of the samples gave spectra close to the calibration samples. Statistical tests were performed by GLM, statistical package Minitab 16. Significant differences ($P<0.05$) between mean values in figures and tables are labelled with different letters. Error bars in the figures indicate standard error of the mean (SEM).

Results and discussion

All tillers of primary growth were in vegetative stage at harvest 2 July and 2 August. At 2 September, a few shoots showed inflorescence, but almost all the tillers were in vegetative stage. Regrowth of harvest regimes A, B, and C was also in vegetative stage. Absence of stem elongation and flowering in *A. flexuosa* is common in boreal forest, probably because of low light intensity (Strengbom *et al.*, 2004).

Grass dry matter (DM) yield and net energy lactation (NEL) yield in undisturbed populations, based on registrations 2 July (A), 2 August (B) and 2 September (D), increased until the last harvest ($P<0.001$) (Figure 1). This shows that production of new tillers and leaf growth throughout the season was larger than leaf and tiller death in the same period. The explanation for this may be that *A. flexuosa* is an inherently slow-growing grass species with a long lifespan of roots and leaves (Poorter and Pothman, 1992; Reich *et al.*, 1992), and with very little stem elongation and flowering. Increasing difference between DM yield and NEL yield throughout the season is due to higher energy value of the plants early in the growing season than later in the season (Figure 1).

Total DM yield was lowest in cutting regime C with three cuts ($P=0.009$). However, total net energy yield showed no statistical significant difference between the harvesting regimes ($P=0.12$) (Figure 2). From these results total energy yield seems to be relatively little affected by harvesting regime, whether harvesting once in late summer or two or three times during the season. Grazing, however, can affect this result as trampling, poaching and tearing of plants come in addition to cutting. Harvesting at different times in the season showed clear differences in growth rate in the different periods. Dry matter yield harvested 2 July (average of cutting regime A and C) accounted for 58% of total yield, while regrowth in the period from 2 August to 2 September, accounted for only 11% in cutting regime B. Coverage of *A. flexuosa* was very high in this experiment with an average dry matter yield for all harvesting regimes of about 800 kg ha^{-1}. This is probably somewhat higher than usual in blueberry forest. Previous surveys in less fertile blueberry forest showed about 250 kg DM ha^{-1} in sparse forest and less than 100 kg DM ha^{-1} in dense forest (Todnem and Lunnan, 2012).

A. flexuosa harvested 2 July had higher energy value and less indigestible fibre than later in the season (Table 1). Reduced energy value and increased content of indigestible fibre during the first part of the growing season, and small changes in the second part, have also been found in previous Norwegian studies (Lunnan and Todnem,

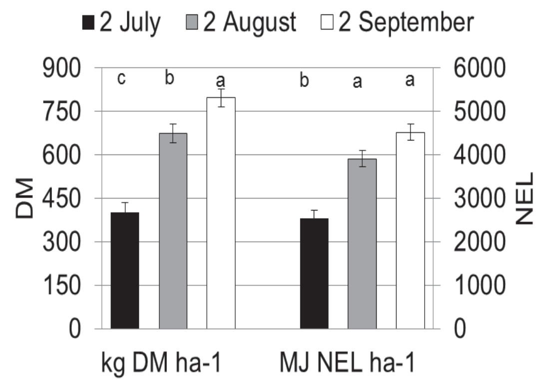

Figure 1. Dry matter yield, kg ha^{-1} and net energy yield, MJ ha^{-1}, in undisturbed population. Average of duplicates, four replicates and two years (n=16).

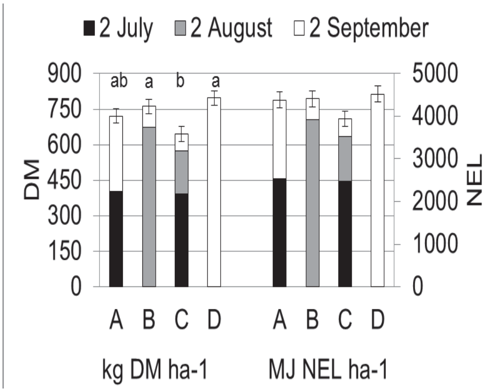

Figure 2. Dry matter yield, kg ha^{-1} and net energy yield, MJ ha^{-1}, at different harvest regimes. Average of duplicates, four replicates and two years (n=16).

Table 1. Energy value (net energy lactation, MJ kg^{-1} DM), indigestible fibre (iNDF, g kg^{-1} DM) and crude protein (CP, g kg^{-1} DM). Average of four replicates and two years (n=8).

Harvest time	NEL, MJ kg^{-1}DM	iNDF, g kg^{-1} DM	CP, g kg^{-1} DM
A – 2 July	6.3 a	88 a	106 a
B – 2 August	5.8 b	107 b	94 a
D – 2 September	5.7 b	114 b	78 b
Regrowth, 2 July to 2 Sept. (A)	5.8 b	109 b	92 a
SEM / *P*-value	0.05 / *P*<0.001	2.4 / *P*<0.001	3.5 / *P*<0.001

2011; Todnem and Lunnan, 2014). The concentration of crude protein declined towards the end of the season (Table 1). Similar results were found previously in similar studies in Vingelen (Todnem and Lunnan, 2014).

Conclusions

Coverage of *A. flexuosa* was very high in this experiment, and dry matter yield averaged for all harvesting systems was about 800 kg ha^{-1}. This is probably somewhat higher than usual in blueberry mountain birch forest. *A. flexuosa* showed great flexibility in relation to defoliation, as grass yields and feed quality were little affected by harvesting management.

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Net replacement film in round bale ensiling of ley crops

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Abstract

Silage quality was compared in round bales where the net was replaced with a film, so called mantelfilm and wrapped with 4, 6 or 8 layers of polyethylene stretch film in a randomized block design with 6 bales per treatment. Bales with mantelfilm had a better shape (lower volume) and were tighter with higher seal integrity and higher CO_2 content, lower mould counts and less ammonia nitrogen. When evaluating the effect of number of stretch film layers it was concluded that more layers gave tighter bales and better silage quality. Four layers of stretch film resulted in inadequate tightness and replacing net with mantelfilm did not seem to bring about the possibility to decrease the number of stretch film layers.

Keywords: silage, round bales, stretch film, mantelfilm, net replacement

Introduction

Prior to stretch film is wrapped on silage round bales to make them air tight, a net is applied at the round surface of the bale in order to maintain the shape. At opening the bales problems occur as the net often entangles to the silage and is difficult to remove. Furthermore net and film have to be removed and sorted separately for recycling. To solve these problems the net can be replaced by a specially developed net replacement film, a mantelfilm. The mantelfilm can be stretched tighter around the bale compared to the net, resulting in bales with stricter cylinder shape in contrast to spherical bales. This makes them more steady and robust when handled. The present experiment was designed to compare the bale shape, the bale integrity and the silage quality when mantelfilm or net was used. One hypothesis is that the mantelfilm will improve seal integrity and anaerobicism of the bale and ameliorate final silage quality. Another hypothesis was also that the use of mantelfilm could reduce the number of stretch film layers needed.

Materials and methods

A mixed ley dominated by timothy (*Pleum pratense*) and some red clover (*Trifolium pratense*) was harvested as first the cut on 6 June 2015 in Sweden (N57°28'; E14°08'). It was fertilized in the spring with 25 Mg cattle slurry and mineral fertilizer (51 kg N, 7 kg S). The crop was cut with a mower conditioner and pre-wilted wide-spread during 24 h. On 7 June the grass was swathed and on 8 June 36 round bales were produced by a McHale Fusion 3 Plus combined baler/stretch film applier (McHale; Ballinrobe, Ireland) that could handle both net and mantelfilm. Six treatments, Net or Mantel film covered with 4, 6 or 8 layers stretch film each, were applied in 6 replicates evenly distributed in 6 blocks over the field. Stretch film of 750 mm width and 25 μm were used (Triowrap; Trioplast AB, Smålandsstenar, Sweden) and mantelfilm was 1390 mm wide and 17 μm pre-stretched film (TrioBale Compressor; Trioplast AB). Samples from one bale per block were taken for chemical analysis of the green crop. Bale weight, height and perimeter at three levels were measured on wrapped bales in the field before moved by a bale grip (Kellfri AB, Skara, Sweden) for transport to storage on a hard surface to be stored under by bird-net. After 17 weeks storage the seal integrity was measured by the vacuum method where a pressure of -200 Pa compared to the atmospheric pressure was applied at each bale and the time until -150 Pa was reached was registered (Spörndly *et al.*, 2008). Seal integrity is expressed in seconds resisting the change in pressure. Before opening the bales carbon dioxide and oxygen was measured with a portable gas analyser GA 2000 (Geotechnical Instruments, Warwickshire, UK) and bales were weighed and size measured. After removing the film visible spots of yeast and mould were measured and 6 drilling cores per bale were pooled into one sample per bale for chemical analyses. Dry matter (DM) and water soluble carbohydrates (WSC) were analysed on complete sample and pH, ammonia nitrogen (NH_3-N), ethanol, lactic acid and acetic acid was analysed on the liquid phase. In the green crop DM, ash, crude protein (CP), neutral detergent fibre (NDF) were analysed and organic matter digestibility (OMD) was calculated from rumen digestible organic matter. All analysis were performed with wet chemistry methods as described by Åkerlind *et al.* (2011).

To calculate the effect of net or mantelfilm and number of stretch film layers on bale volume, perimeter, density, DM losses, CO_2, seal integrity, yeast area, mould area, pH, WSC, ammonia, lactic acid, acetic acid and ethanol the PROC GLM in the SAS package (SAS Institute Inc, Cary. NC. USA) was used for the following model:

$$Y_{ijkl} = \mu + \text{Mantel/Net}_i + \text{Layer}_j + \text{Mantel/Net}\times\text{Layer}_k + \text{Block}_l + \varepsilon_{ijklm}$$

where $_i$ is mantelfilm or net, $_j$ is 4, 6 or 8 layers of strech film, $_k$ is the interaction between Mantel/Net and Layer, $_l$ is the block and ε_{ijklm} is the residual effect.

The effect was considered significant when P<0.05.

Results and discussion

The composition of the crop indicates a high quality ley with 148 g CP kg^{-1} DM, 521 g kg^{-1} DM and OMD 797 g kg^{-1}. Average DM content of the silage was 445 g kg^{-1} DM. Average weight of the bales was 662 kg fresh matter. Bales were slimmer with smaller perimeter when mantelfilm was used resulting in smaller volume and a tendency for higher density (Table 1). Mantelfilm bales had better seal integrity expressed in longer time resisting the change in pressure and higher CO_2 content in the bales. Bales with better tightness are more resistant to the entrance of oxygen which is essential for preventing mould growth and consequently, the mantelfilm bales had less mouldy area on the surface after opening. No difference in pH and lactic acid was detected between treatments. Higher ammonia nitrogen and a tendency for lower WSC in the netted bales suggests that mould and other bacteria than lactic acid bacteria have used oxygen leaking into these bales for protein degradation.

The effect of number of stretch film layers on the shape of the bales was indicated by a wider perimeter when more layers were applied. The force of the stretch film shapes the cylindrical bale to becoming more round and more layers represent a higher force. However, no effect of layer number was seen on volume or density. More layers increased the CO_2 content inside the bales and the seal integrity with only 4 layers was very low. The effect of low seal integrity associated with fewer layers was more mould and less WSC. No changes in pH was evident but the ethanol content clearly increased, although by low numbers, when stretch film was applied with less than 8 layers. Ethanol is often associated with yeast growth but yeast was however hardly detected at all, except to a marginal extent in the 4-layer bales.

Interaction between mantelfilm/net and number of layers came out significant for the following variables: CO_2, seal interity, mould, WSC, pH, lactic acid and acetic acid. Negative effects, such as low seal integrity and elevated mould counts, connected to fewer layers were more pronounced in bales with net compared to bales with mantelfilm. However, at the lowest number of layer also mantelfilm bales expressed unsatisfactory low seal integrity of 234 seconds.

Table 1. Least square means and SEM from the model comparing mantelfilm with net and 4, 6 or 8 layers of stretch film (36 observations). Different superscripts in the same row indicate significant contrast differences at $P<0.05$.

	Comparing Mantelfilm vsNet			Comparing number of stretch film layers				Mantel × layer[1]
	Mantelfilm	Net	SEM	4 layers	6 layers	8 layers	SEM	
Volume, m^3	1.67^a	1.71^b	0.009	1.69	1.69	1.68	0.010	n.s.
Perimeter, m	4.21^a	4.28^b	0.013	4.22^a	4.24^{ab}	4.28^b	0.016	n.s.
Density, kg DM m^{-3}	172.7	167.6	2.50	170	170	171	3.0	n.s.
DM loss, %	0.90	0.90	0.126	0.96	0.95	0.80	0.155	n.s.
CO_2, %	63.7^a	57.2^b	1.73	54.0^a	61.3^b	66.1^b	2.12	$P<0.05$
Seal integrity, s	938^a	533^b	137.2	165^a	879^b	1162^b	168.0	$P<0.05$
Yeast, cm^2	0.00	0.06	0.039	0.09	0.00	0.00	0.048	n.s.
Mould, cm^2	0.03^a	0.78^b	0.224	1.17^a	0.06^b	0.00^b	0.275	$P<0.05$
pH	5.3	5.3	0.03	5.3	5.3	5.3	0.04	$P<0.05$
WSC, g kg DM^{-1}	7.2	6.4	0.39	6.0^a	6.9^{ab}	7.5^b	0.48	$P<0.05$
Ammonia-N, % of total	4.5^a	5.1^b	0.21	5.1	4.6	4.7	0.26	n.s.
Lactic acid, g kg DM^{-1}	1.4	1.5	0.13	1.6	1.3	1.4	0.16	$P<0.05$
Acetic acid, g kg DM^{-1}	0.4	0.4	0.029	0.4	0.3	0.4	0.03	$P<0.05$
Ethanol, g kg DM^{-1}	1.5	1.6	0.058	1.8^a	1.6^b	1.3^c	0.07	n.s.

[1] Mantel × layer indicates the interaction between mantel film or net and number of layers where n.s. = not significant.

Conclusions

Round bales where mantelfilm was used expressed a more cylindrical shape, had a better seal integrity and better silage quality than bales where net was used. More layers of stretch film resulted in better seal integrity and better silage quality. Four layers of stretch film resulted in inadequate bale tightness of the bales, also when mantel film was used.

Acknowledgements

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Modelling gas production from silage fermentation

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Abstract

The aim of this study was to evaluate models fitting gas production data from silage fermentation. A semi-automated laboratorial system was used for measuring gas production during the fermentation of maize, sugarcane, elephant grass and reconstituted corn grain silages for 160 days. Five 1.96-L silos were used for each substrate. Data were recorded every 12 h until d-3, every 24 h from d-4 until d-21, every 3 d from d-22 until d-30 and every week until the end of the storage, and computed as cumulative gas production per kg DM. Twelve models (without lag) were fitted to data, using the NLIN procedure of SAS: exponential 1- and 2-pools (EXP and EXP2), logistic 1- and 2-pools (LOG and LOG2), Groot 1- and 2-pools (GROOT and GROOT2), Weibull 1- and 2-pools (WEIB and WEIB2), Gompertz 1- and 2-pools (GOMP and GOMP2), and France 1- and 2-pools (FRANCE and FRANCE2). Model adequacy was dependent on crop. However, all 1-pool models converged and had relatively high R2 (≥0.90) and low root mean squared error (RMSE), satisfying the minimum criteria for a good model. Overall, the goodness of fit was improved with 2-pools, but several 2-pool models did not converge. In conclusion, relatively simple models such as a single exponential model would be used to fit gas production from silage fermentation.

Keywords: anaerobic fermentation, carbon dioxide, kinetic, volume

Introduction

During silage fermentation, gases are formed as by-products of microbial metabolism and represent the main sink of dry matter (DM) loss. Therefore, measuring gas production kinetics is a method to determine microbial activity and nutrient losses. Although few studies have investigated the gas production in silages (Honig, 1968; Meiering *et al.*, 1968), at our knowledge, there is no report comparing models to describe the kinetics of gas formation during silage storage. Thus, the aim of this study was to evaluate models fitting gas production data from silage fermentation.

Materials and methods

A semi-automated laboratorial system (Daniel and Nussio, 2015) was used for measuring gas production during the fermentation of maize, sugarcane, elephant grass and reconstituted corn grain silages for 160 days. Five 1.96-L silos were used for each substrate. At ensiling, mass porosity (Pitt, 1986) was equalized at 0.40 for whole-plant crops (maize, sugarcane and grass) and at 0.20 for reconstituted corn grain. The internal pressure of the silos was measured by using a pressure transducer (DataPress, MPL, Piracicaba, Brazil). Readings were taken every 12 h until d-3, every 24 h from d-4 until d-21, every 3 d from d-22 until d-30 and every week until the end of the storage. Afterwards, pressure was converted to volume and computed as cumulative gas production per kg DM (see Daniel and Nussio, 2015). Twelve models without lag phase were fitted to data, using the NLIN procedure of SAS (v. 9.3, SAS Institute Inc., Cary, NC): exponential with 1- and 2-pools (Exp and Exp2), logistic with 1- and 2-pools (Log and Log2), Groot with 1- and 2-pools (Groot and Groot2), Weibull with 1- and 2-pools (Weibull and Weibull2), Gompertz with 1- and 2-pools (Gomp and Gomp2), and France with 1- and 2-pools (France and France2). Goodness of fit were compared by R^2-adjusted (largest is better) and root mean squared error (RMSE) (smaller is better).

Results and discussion

Model adequacy was dependent on crop. However, all 1-pool models converged and had relatively high R^2 (≥0.90) and low RMSE (Table 1), satisfying the minimum criteria for a good model. Overall, the goodness of fit was improved with 2-pools, but several 2-pool models did not converge, especially Groot2, Weibull2 and France2. In addition, although the non-first-order models fit the data better than simpler models (e.g. exponential with 1 pool), their parameters do not always permit unequivocal comparison of substrates (Huhtanen *et al.*, 2008). Therefore, simpler models such as a single exponential model may be adopted to describe the kinetics of gas formation during silage storage (Figure 1).

Table 1. Fit statistics of 1- and 2-pool models describing gas production during fermentation of selected silages.[1]

	Model											
Silage	Exp	Log	Groot	Weibull	Gomp	Fran	Exp2	Log2	Groot2	Weibull2	Gomp2	Fran2
	R^2-adjusted											
Maize	0.99	0.96	0.99	0.86	0.96	0.98	0.99	0.99	NC	NC	0.99	NC
Sugarcane	0.98	0.98	0.98	0.99	0.98	0.99	0.99	0.99	0.99	0.99	0.99	0.99
Elephant grass	0.94	0.92	0.99	0.99	0.90	0.99	0.99	0.98	NC	NC	0.98	NC
Reconstituted corn grain	0.94	0.92	0.99	0.97	0.91	0.99	0.99	0.98	NC	NC	0.99	NC
	RMSE (L/kg DM)											
Maize	0.09	0.19	0.05	0.13	0.19	0.13	0.02	0.06	NC	NC	0.04	NC
Sugarcane	1.10	0.94	0.95	0.83	1.21	0.78	0.49	0.59	0.25	0.80	0.36	0.24
Elephant grass	0.22	0.20	0.03	0.04	0.27	0.03	0.05	0.11	NC	NC	0.10	NC
Reconstituted corn grain	0.54	0.59	0.22	0.32	0.67	0.26	0.12	0.28	NC	NC	0.22	NC

[1] NC: model failed to converge.

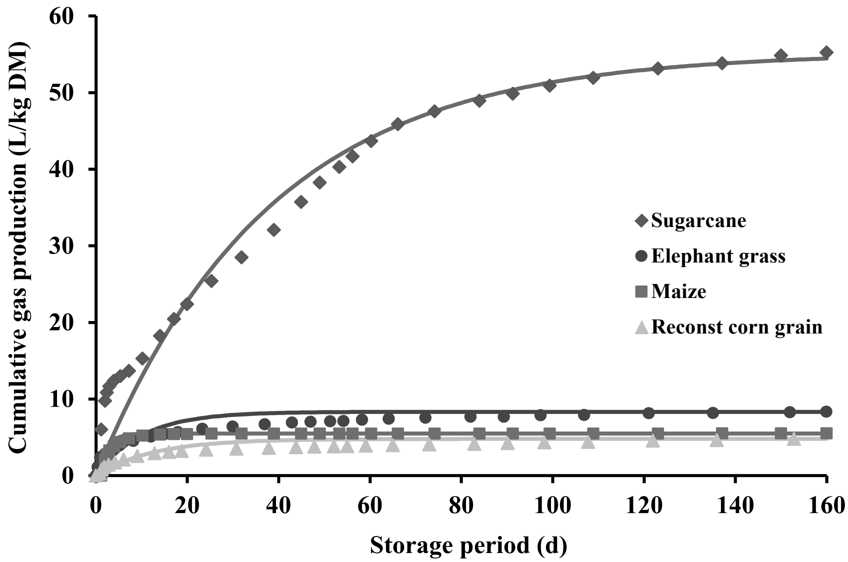

Figure 1. Cumulative gas production fitted with exponential 1-pool model (solid lines) in selected silages.

Conclusions

Relatively simple models such as a single exponential model may be used to fit gas production from silage fermentation.

Acknowledgements

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Optimization of the harvesting time of pure lucerne (*Medicago sativa* L.) swards in Finland

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Abstract

To reveal the optimal harvesting time of pure lucerne (*Medicago sativa L.)* swards in Finland, the sward development, dry matter (DM) yield and feeding value of four cultivars (Alexis, Lavo, Live, Nexus) were investigated at Luke Ylistaro, Finland. Before the first cut, the developmental rate of cultivars was similar; however, their growth maxima peaked at different time and maximum leaf area indices were reached at 180 °Cd. Before the second cut, rates and maxima of plant growth differed between cultivars until 210 °Cd, but leaf area indices peaked at 350 °Cd for all cultivars. During the third cut, plant growth was slow and leaf area indices were still increasing at harvest (at 330 °Cd). DM yields of cultivars were different only in the third cut when DM yields of Alexis and Live were 25% higher than those of Lavo and Nexus. D-values of cultivars were similar but below the desired level for silage. In Finland, pure alfalfa should be harvested during the early or late bud stage. D-value and iNDF may be predicted by the height and developmental stage of the sward.

Keywords: *Medicago sativa* L.*,* tiller growth, leaf area index, harvesting time, feeding value

Introduction

Recently, there has been an increasing interest towards growing lucerne (*Medicago sativa L.*) for forage in Finland. There are no advisory frames for what is the optimal harvesting time of pure lucerne stands in Nordic conditions. In other countries, lucerne is recommended to be harvested at late bud stage or at early blooming (Myers and van Keuren) or when a certain effective temperature sum has been accumulated (Kaatz, 2015). In this study, the developmental processes of a pure lucerne sward during the first harvest year were examined.

Materials and methods

A field study with lucerne (cv. Alexis, Nexus, Live and Lavo) is being carried out at Luke Ylistaro, Finland (62°95'N 22°53'E) during 2014-2016. Plots were established in 2014 in four replicates on a sandy silt (pH 6.3) using 700 seeds m^{-2} and 30, 3 and 3 kg ha^{-1} of N, P and K, respectively. During 2015 plots were fertilized in the spring with NPK 40-4-4 kg ha^{-1}, for second cut with NPK 33-10-41 kg $ha^{-1}$and for third cut NPK 20-0-38 kg ha^{-1}. Weekly, plots were measured for leaf area index (LAI; LAI-2000 Plant Canopy Analyzer, LI-COR Inc., Lincoln, Nebraska, USA) and for stretched height (mm above ground level) and developmental stage (BBCH scale, Lancashire *et al.,* 1991) of main and side tillers of randomly marked plants (n=16 per cv.). Data and shoot growth (difference between each measuring time) were contrasted to the accumulated effective temperature sum (°Cd; base +5 °C, the zero point of °Cd at the beginning of the growing season). Cuts were taken at late bud stage (16 June 2015), at the beginning of blooming (5 Aug 2015) and when the growth had ceased (9 Sept 2015). Samples were dried 24 h at 60 °C for dry matter (DM) content and analyzed for feeding values with NIRS (legume calibration, Valio Ltd., Seinäjoki, Finland). Data were analyzed using the MIXED Procedure of SAS (version 9.4; SAS Enterprise Guide 7.1, SAS Institute Inc., Cary, NC, USA) having cultivar as fixed and replicate as random effect (P<0.05). Correlations were analyzed with the REG Procedure.

Results and discussion

LAI developed similarly in all cultivars during the growth of first and third cut sward (Figure 1). In the second cut sward, the lag phase of tiller growth after the cut lasted nearly until 150 °Cd, and the increase of LAI was more moderate. LAI continued to increase till the second cut in Lavo, while LAI values decreased in other cultivars after 350 °Cd. The maxima of LAI in the third cut sward were clearly lower than in the swards of previous cuts.

Growth rate (mm °Cd^{-1} $tiller^{-1}$) of cultivars did not differ in the first cut sward. Growth rates were high, 4-10 mm °Cd^{-1} $tiller^{-1}$ before a clear decrease in growth at 150 °Cd (Figure 2). In the second cut sward, Lavo had the highest growth rates until 140 °Cd. Rates were, however, lower than in first cut sward (1-7 mm °Cd^{-1} $tiller^{-1}$). From 140 to 210 °Cd of regrowth, Alexis had the highest growth rate but the lowest LAI, which was probably due to low tiller density (Table 1). Cultivars had diverse recovery abilities: Lavo is not easily disturbed by cutting, while other cultivars need more time for regrowth. In the third cut sward there were no differences and growth rates were low (2-4 mm °Cd^{-1} $tiller^{-1}$).

Cultivars differed in DM yields only in the third cut, as Alexis and Live had the highest yields (Table 1). Constantly, tiller densities were the highest in Lavo and lowest in Alexis. This may have affected LAI values (Figure 1). Feeding values did not differ between cultivars within different cuts. D-values

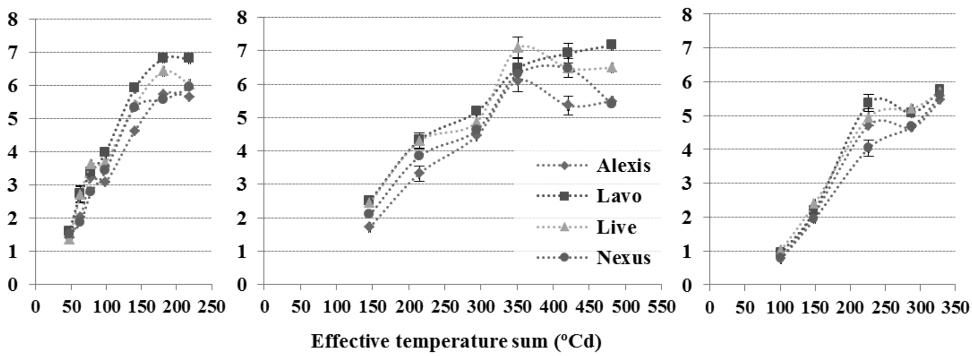

Figure 1. Development of leaf area index (LAI) in pure swards of lucerne during first, second and third cut sward in relation to the accumulated effective temperature sum (°Cd; above base temperature +5 °C). Standard error of mean (SEM) is given by error bars, if difference between cultivars was significant (P<0.05).

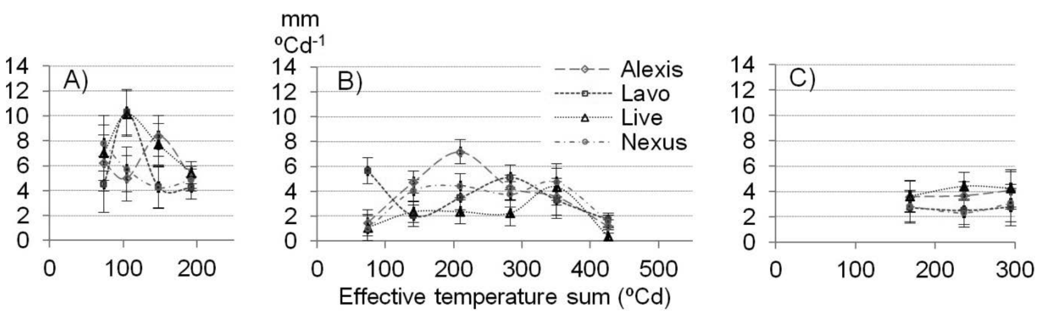

Figure 2. The rate of plant growth (mm °Cd^{-1}) of individual lucerne plants (including main tiller and all side tillers up to 11th side tiller) in the first cut sward (A), second cut sward (B) and third cut sward (C).

(concentration of digestible organic matter in DM) were 669, 594 and 635 g kg^{-1} DM in first, second and third cut, respectively, which are below the desired level (680-700 g kg^{-1} DM) for forages in Finland. In a recent meta-analysis of Lucerne, the same level of digestibility in the regrowth was reported when the cutting regime was 9 weeks (Baumont *et al.,* 2014). In this study, the concentration of indigestible fibre (iNDF) was high: 117, 177 and 144 g kg^{-1} DM in first, second and third cut, respectively. Concentration of protein was ca. 200 g kg^{-1} DM in the first and the second cut and 260 g kg^{-1} DM during the third cut, corresponding with results when cutting at the same developmental stage (Baumont *et al.,* 2014). The concentration of reducing sugars was low (105, 72 and 52 g kg^{-1} DM), which may result in problems in ensiling (Vuorinen and Takala, 1993).

LAI values, reflecting the DM yield potential, reach their maxima during bud stage showing this would be the optimal cutting time of pure lucerne. At the same time, digestibility is expected to be good. Based on LAI measurements the optimal cutting regime may be 5 to 6 weeks after the first cut, supporting the observations by Baumont *et al.* (2014), but there may be variation between cultivars due to differences in LAI (Figure 1). The height and BBCH correlated negatively with D-value (height R^2=0.79 and BBCH R^2=0.83) and positively with iNDF (height R^2=0.77 and BBCH R^2=0.82). Thus, measuring BBCH or the height of the sward may apply as a simple tool to evaluate the digestibility of lucerne swards.

Table 1. Dry matter (DM) yield and tiller density in the first, second and third cut of lucerne during 2015 at Luke Ylistaro, Finland. Pairwise differences are shown by letters (a, b, c).

	Cut 1				Cut 2				Cut 3			
	DM yield, kg ha^{-1}		Tiller density, m^{-2}		DM yield, kg ha^{-1}		Tiller density, m^{-2}		DM yield, kg ha^{-1}		Tiller density, m^{-2}	
Alexis	3,297	a	632	a	3,890	a	705	a	2,098	a	778	a
Lavo	3,117	a	1,327	c	3,707	a	1,004	c	1,535	b	1,142	b
Live	3,573	a	1,046	b	4,447	a	937	bc	2,067	a	987	ab
Nexus	3,472	a	1,022	b	3,685	a	834	ab	1,717	b	850	a
average	3,365		1,007		3,932		870		1,854		939	
SEM	187.9		84.9		316.9		97.8		98.1		91.4	
P	ns		<0.001		ns		<0.05		<0.01		<0.05	

Conclusions

It is possible to harvest high DM yield with good digestibility in pure lucerne swards when the timing of harvest is at early or late bud stages. For regrowth, cutting regime may be 5 to 6 weeks, depending on the cultivar. The measurement of height and developmental stage of the sward may apply as easy tools to predict the changes in the D-value.

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Variability of forage quality between and within three maturity groups of *Lolium perenne* L. during the first growth

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Abstract

A set of 39 different cultivars of *Lolium perenne* (Lp) assigned to early, medium and late maturing types were harvested and analysed at weekly intervals during the first growth. Significant differences between the heading groups were found with a time lag of 6 days for crude protein content resp. up to 11 days for digestibility of organic matter and energy concentration. The early heading cultivars strongly differed from the other two groups, which performed comparably and more resilient for a longer time. Within all three heading groups a strong variation could be noticed, leading to some unexpected overlapping. Our results indicate that the assignment of Lp-cultivars to the heading groups is imprecise and should be improved by advanced recordings resp. analyses during the time of the official variety testing period. This may lead to a more sufficient selection of Lp-cultivars for grassland mixtures and therefore increase the quality of home-grown forage.

Keywords: *Lolium perenne*, maturity groups, persistence, variety

Introduction

To improve the quality of home grown forage from grassland, different strategies have been developed. Among others the establishment and renovation of grassland by over-seeding with high-quality seed mixtures are of great importance. Seed mixtures for grassland are containing well tested cultivars of forage grasses and legumes. *Lolium perenne L.* (Lp) is one of the most common grasses used in seed mixtures for meadows and pastures all over Europe and there is a huge number of cultivars with different quality characteristics available (Krautzer *et al.*, 2013; McDonagh *et al.*, 2015). The selection of cultivars for seed mixtures depends on site conditions, type of grassland management (utilisation frequency, fertilization intensity) and on the intended purpose (e.g. establishment or over-seeding, meadow or pasture). Lp-cultivars are classified into early, medium and late maturity groups (based on heading dates) however early heading cultivars are not necessarily also early maturing. There is still little information available on the variation of quality parameters between and within these groups. Therefore the objectives of our project were: (1) to illustrate the dynamics of quality parameters during the first growth; (2) to identify the variation between and within the maturity groups; and (3) to determine the optimal harvesting time. The findings will both support the selection of suitable Lp-cultivars for seed mixtures under changing environmental conditions and provide grassland farmers with useful information concerning forage quality (Reheul *et al.*, 2013; Blackmore *et al.*, 2015).

Materials and methods

Following the official variety testing period of three years, we harvested 39 cultivars of Lp assigned to three maturity groups in weekly intervals during the first growth. The field experiment was conducted at the Federal Research and Education Centre Raumberg-Gumpenstein, Austria (47°29`40``N and 14°06`11``E, 700 m a.s.l.) with an average annual precipitation of 1,015 mm and a long-term mean annual air temperature of 8.4 °C. Yield, forage quality, especially crude protein (CP; VDLUFA, 1976) content, digestibility of organic matter (dOM; Tilley and Terry, 1963) and energy concentration (NEL; calculated according GfE, 2001) were determined to find out the variability between and within the maturity groups. Analyses of variance (ANOVA) were performed with the software package SPSS version 22.0 (IBM®SPSS®Statistics), followed by a multiple range test (Bonferroni post hoc test) using a significance level of $P<0.05$.

Results and discussion

The self-supply of energy and protein has become an important issue in European agriculture and therefore there is still an increasing interest to improve forage quality on grassland farms. Our results clearly confirm the decreasing protein concentration with delayed harvest time during the first growth showing a linear mean drop of 15 g CP kg^{-1} DM per week for all three maturity groups (Figure 1). On average the late heading Lp-cultivars performed best, followed by the medium heading cultivars and by the early heading group with significantly lower CP-concentrations at most of the sampling dates. A concentration of 150 g CP kg^{-1} DM which is at least required for high yielding cows was achieved on 14 May for the early heading group, on 17 May for the medium heading group and again three days later on 20 May for the late heading group. Even though there seems to be a clear differentiation between the three groups, a strong variation within the groups could be noticed with some remarkable overlapping. Two cultivars of the early heading group for example had much higher CP-concentrations that were comparable with those of the late heading group. This finding could also be used to select early heading cultivars providing good winter hardiness and high forage quality as well.

The dynamic of dOM during the first growth was following a polynomial function with a clear shift between the three maturity groups. In the early heading group the maximal dOM was achieved on 18 May with a decrease of 3% per week afterwards, whereas the highest dOM of both other groups was obtained 7 resp. 11 days later, remaining at a high level for another week. Again some early heading cultivars performed very similar to the medium and late heading group. The energy concentration (expressed in MJ NEL kg^{-1} DM) was strongly linked with dOM and therefore the dynamic during the observation period but also between the three maturity groups was very similar (Table 1).

For farmers, not only high forage quality but also sufficient yield is of great importance. Our results indicate that there is a relatively small time span to achieve high forage quality with early heading Lp-cultivars in combination with high yield in the first growth. In contrast medium and late heading Lp-

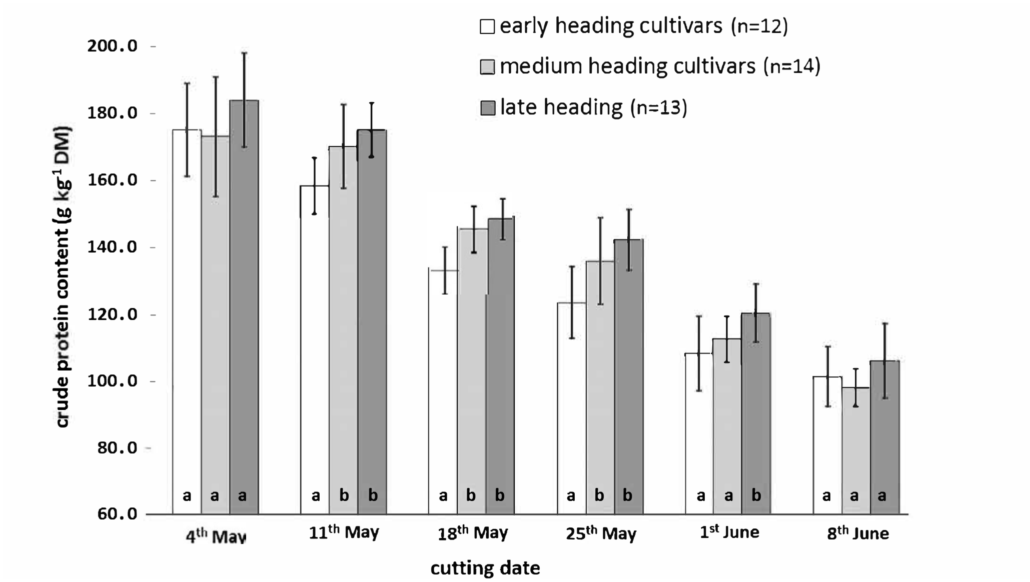

Figure 1. Crude protein content of *Lolium perenne* cultivars in the course of the first growth.

Table 1. ANOVA for selected parameters of forage quality assigned to heading groups (early, medium, late) and six weekly harvest intervals (from 4 May to 8 June) in Austria.

	Dependent variable[1]		
	CP (g kg^{-1} DM)	dOM (%)	NEL (MJ kg^{-1} DM)
Heading group (Hg)	sign.	n.s.	n.s.
Harvest date (Hd)	sign.	sign.	sign.
Hg × Hd	n.s.	sign.	sign.
R^2	0.88	0.46	0.37

[1] Significance level $P<0.05$.

cultivars performed much more resilient and provide a later and extended time slot of about 10-12 days for high forage quality and sufficient yield.

Conclusions

It has become very common to use a set of differently heading Lp-cultivars for grassland seed mixtures to provide a larger harvest window and to reduce the risk of unfavourable weather conditions, which in mountainous regions very often occur during the harvest time of the first growth. As there is a strong variability of forage quality between and within the maturity groups of Lp-cultivars more specific information about these characteristics are desirable.

There is an ongoing discussion in Austria to implement more detailed recordings and analyses during the official, three years lasting variety testing period which should provide more reliability in the selection of Lp-cultivars for grassland seed mixtures.

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Alkaloid content variability in *Lolium perenne* infected with *Epichloë* endophytes in natural grasslands

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Abstract

Many grasses are asymptomatically infected by *Epichloë* fungal endophytes. Infected plants may contain diverse fungal alkaloids, some of them are toxic to mammals (ergovaline, lolitrem B), and others are insect feeding deterrents (peramine). The alkaloid profile of infected plants depends on factors such as the plant and fungal genotype and environmental conditions. In this work we studied the alkaloid content of a heterogeneous set of *Lolium perenne* plants from natural grasslands, infected with different morphotypes of *Epichloë* endophytes. The most frequent alkaloid was peramine, found in 77.4% of the plants, followed by lolitrem B (67.7%) and ergovaline (62.7%). The concentration of alkaloids varied depending on the *Epichloë*-morphotypes hosted: most of plants infected by the M3 morphotype had lolitrem B, M1-infected plants had the highest ergovaline content, and M2-infected plants had the highest peramine concentration. Several plants without alkaloids toxic to mammals, but with peramine, were detected. These were infected with the M2 stroma-forming endophyte *Epichloë typhina*.

Keywords: Lolitrem B, ergovaline, peramine, perennial ryegrass, *Epichloë* endophytes

Introduction

Epichloë fungal endophytes are symbionts of several grass species from temperate zones (Saikkonen *et al.,* 2015). Infected plants are known to contain several mycotoxins and three are particularly important in *Lolium perenne*: lolitrem B, ergovaline and peramine (Bush *et al.*, 1997). Lolitrem B is a tremorgenic compound responsible for ryegrass staggers, a toxic syndrome affecting mainly sheep. Ergovaline is an ergopeptine alkaloid causing various diseases in cattle, like fescue-foot and fescue toxicosis. Peramine is an alkaloid toxic for insects but not for mammals. The alkaloid profile of infected plants depends on factors such as the plant and fungal genotype and the surrounding environmental conditions (Lane *et al.*, 2000; Schardl *et al.*, 2012). In a previous study we detected several types of *Epichloë* morphotypes in natural populations of *L. perenne* (Soto Barajas *et al.*, 2013). The objective of this work was to analyse the variability in alkaloid contents of a heterogeneous set of *L. perenne* plants from natural grasslands, infected with different morphotypes of *Epichloë* endophytes.

Materials and methods

Ryegrass plants were collected at six wild grasslands with different ecological conditions located in the west of Spain. Infection with *Epichloë* endophytes was verified by isolation of the fungus from stem and leaf sheath fragments in potato dextrose agar (PDA) (Bacon and White 1994). Some plants were infected by the symptomatic stroma-forming endophyte *Epichloë typhina*. The endophytes isolated from asymptomatic plants were classified into three morphotypes based in their morphological characters observed in PDA (M1, M2 and M3) (Soto Barajas *et al.*, 2013). All plants were transplanted in autumn to a plot in an experimental farm (Salamanca, Spain), were watered during their establishment but not thereafter, and were not fertilized. The presence of *Epichloë* endophytes and their morphotypes were known for each plant. In May the following year after transplant, the ryegrass was harvested at the flowering stage by cutting the aboveground biomass. Plant samples were freeze-dried and ground for chemical analyses. Chemical analyses of the alkaloids lolitrem B, ergovaline and peramine were performed by high performance liquid chromatography (HPLC). Lolitrem B was analysed following the method of Gallagher *et al.* (1985), ergovaline was analysed according to the procedure described by Yue *et al.* (2000) and peramine with slight modifications of the methods of Barker *et al.* (1993) and Yue *et al.* (2000). This three alkaloids were analysed in all endophyte-infected plants (n=85) and in 15 non-infected plants randomly selected. Since the distribution of *Epichloë*-morphotypes was not equal in all sampled grasslands and only in three of them the four morphotypes occurred, the effects of morphotype and plant origin on alkaloid contents were analysed separately by means of a one-way ANOVA.

Results and discussion

Fungal alkaloids were present in endophyte-infected plants but not in plants without *Epichloë*. Lolitrem B was detected in 67.7% of plants, ergovaline in 62.7% and peramine in 77.4% of the *Epichloë*-infected plants. The highest proportion of plants with lolitrem B was found in those infected with *Epichloë* M3-morphotype and the lowest in M2-infected (Table 1). Similarly, the highest concentration of lolitrem B was detected in M3-infected plants. The proportion of plants with ergovaline ranged from 22.2% in plants with M2-stroma-morphotype to 84.6% in M3-infected plants, but differences in the concentration of this alkaloid among *Epichloë*-morphotypes were not statistically significant (Table 1). Peramine was detected in 42.8% of M2-infected and in 92.0% of M3-infected plants. The highest peramine concentration was found in plants infected by M2 morphotypes, including those bearing stromata (Table 1). These results show a strong influence of *Epichloë* morphotype on the concentration of alkaloids in *L. perenne* plants.

We found significant differences in lolitrem B and peramine contents among grasslands. These differences were related to the distribution of *Epichloë* morphotypes in the populations, because the proportion of each morphotype was not equal in the six grasslands (Soto Barajas *et al.,* 2013). For instance, the highest peramine concentration was found in samples from the CR population, where the proportion of M2-stroma type of endophytes was the greatest.

Toxic levels of lolitrem B for livestock (>1.80 mg kg^{-1}) were detected in a few plants, and most of them were infected with M3-endophytes; whereas an important proportion of the plants (80.0%) had a concentration of ergovaline above the reported safe limit for livestock consumption (0.40 mg kg^{-1}). However, reports of livestock intoxications in Europe are scarce mainly because of the high floristic diversity in non-cultivated grasslands and the use of endophyte-free ryegrass cultivars (Zabalgogeazcoa and Bony, 2008). The three alkaloids, peramine, lolitrem B and ergovaline in the same plant sample were detected in ryegrass harboring any of the morphotype of *Epichloë.* This was frequent in plants infected with M3 endophytes (>75.0%), and less frequent in plants infected with M2-endophytes (9.1%) or in M1-infected plants (27.6%). Plants containing only peramine (insecticide) and not the toxic alkaloids lolitrem B or ergovaline were 41.7% of the plants with M2-stroma forming endophytes (*E. typhina*). This

Table 1. Fungal alkaloid concentrations (mg kg^{-1} DM) in *Lolium perenne* plants from grasslands: percentage of plants containing the alkaloid and concentration according to the morphology of the *Epichloë* endophyte.[1]

Epichloë morphotype	% plants with lolitrem B	Lolitrem B (mg kg^{-1})	% plants with ergovaline	Ergovaline (mg kg^{-1})	% plants with peramine	Peramine (mg kg^{-1})
M1	67.3	1.12±0.08 ab	59.6	1.41±0.21	73.1	3.73±0.23 a
M2	57.1	0.89±0.21 ab	57.1	1.08±0.60	42.8	10.11±5.22 b
M3	80.0	1.38±0.11 a	84.6	1.01±0.16	92.0	6.26±0.57 b
M2-stroma	44.4	0.48±0.03 b	22.2	0.10±0.00	88.8	17.52±1.52 c
Total/mean	67.7	1.17±0.06	62.7	1.16±0.18	77.4	6.25±0.60

[1] a,b,c in each column, means with different letters were statistically different ($P<0.05$).

is in agreement with Leuchtman *et al.* (2000) who found that a clear tendency for plants associated with stroma-forming *Epichloë* species to be free of alkaloids, and those that did produce alkaloids contained only small levels of peramine. None of the seed-transmitted infected plants produced peramine alone.

Conclusions

Alkaloids produced by *Epichloë* endophytes (lolitrem b, peremine and ergovaline) are common in *L. perenne* plants from natural grasslands. The concentrations varied depending on the *Epichloë*-morphotype: M2-infected plants had the highest peramine and ryegrass infected with M3 morphotype the highest lolitrem B.

Acknowledgements

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Morphological and productive aspects of sorghum intercropped with legumes

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Abstract

This study reports the response on morphological and yield production of intercropping of sorghum with legumes in comparison to the sorghum monocrop, during the dry season. The treatments tested were dwarf sorghum (cv. Surgo) with soybean (*Glycine max* L. var. Mitsuko) and giant sorghum (cv. Sweet Virginia bmr) with common beans (*Phaseolus vulgaris* L. var. Neckargold), under three different legume seeding rates (7.5, 15 and 30 seeds per m^2), with four replicates. The number of plants established for both cv of sorghum were reduced when in association with legume. At legume seeding rate of 30 seeds per m^2 both sorghum cv increase the stem diameter and leaf production and increased the leaf:stem ratio. Furthermore, the association reduced the giant sorghum height. The intercropping decreased the height and increase the bending inclination of giant sorghum. The intercropping of dwarf sorghum with soybean increase the production of grain/panicle but did not change the forage mass production. The intercropping of giant sorghum with beans decreased the productivity. The intercropping of sorghum with soybean at seeding rate at 15 seeds per m^2 is an alternative to increase productivity and quality of forage, without compromising the morphological development of sorghum.

Keywords: bean, forage, productivity, quality, soybean, sustainable systems

Introduction

In sustainable grassland systems, cultivate sorghum (*Sorghum bicolor* L), in replacement of corn (*Zea mays* L.), has excelled, especially in regions with limited water resources and low fertility soils. This is because sorghum is less sensitive to water stress and low soil fertility (Goufichon *et al.*, 2010). Although lower forage quality is often claimed for sorghum, this issue could be overcome by associating sorghum and legumes (Da Silva *et al.*, 2015). Intercropping compatible species with different niches of nutrient absorption and simultaneous development, always provide favourable results in terms of productivity and ecosystem quality (Koohi *et al.*, 2014). In this context, the objective was to evaluate the effects of specific associations between sorghum and legumes, in three seeding rates, on the DM yield and morphological aspects of the sorghum, in comparison with sorghum in monoculture.

Materials and methods

Field plots (5×1.5 m) of sorghum (*Sorghum bicolor* L) in monocrop or intercropped with legumes were sowed at the INRA Station in Lusignan, during the summer of 2013, in a clayey silt soil of medium fertility. The associations were dwarf sorghum (cv. Surgo) with soybean (*Glycine max* L. var. Mitsuko) and giant sorghum (cv. Sweet Virginia bmr) with common beans (*Phaseolus vulgaris* L. var. Neckargold), under three seeding rates for legumes (7.5, 15 and 30 seeds per m^2) in a randomized complete design, with four replicates. Seeds were sowed in alternate rows, at 22 cm spacing. The sorghum seeding rate was 20 seeds per m^2. Two days after sowing, 61 kg N ha^{-1} were applied and the phytosanitary control was performed by manual weeding. Forage was harvested when the sorghum reached the physiological maturity (dry matter content around 300 g kg^{-1} FM). Immediately before harvesting, sorghum bending index was evaluated by counting the number of broken and/or bended plants per plot. Afterwards, four sorghum plants were collected per plot (16 plants per treatment) for evaluation of height, stem diameter (using a digital caliper) and plant weight, as well as the morphological fractions (stem, leaves+sheath and grains/panicle). Forage yield per hectare was estimated by clipping 2 m linear to the central lines of each plot at 15 cm stubble height, so representing a sample area of 0.6 m^2 per plot. Data were submitted to the analysis of variance using the PROC GLM of SAS®, version 9.3. Treatment means were compared by Tukey test (P<0.05).

Results and discussion

Intercropping with legumes decreased (P<0.05) the number of sorghum established plants, with more evidence at 7.5 seeds of legume per ha, showing values of 196.0 vs144.9 and 210.2 vs 139.2 thousand plants per ha for dwarf sorghum and giant sorghum, respectively (Table 1). This low sorghum population probably occurred because the ecosystem balance, since it may have occurred a competition between sorghum and legumes for nutrients and light. The giant sorghum presented greater height (P<0.05) in monoculture or with low legume seeding rate (7.5 seeds per ha). In these conditions, the reduction in giant sorghum height reached a level equal to dwarf sorghum, in treatments with legume seeding rate high or medium. According to Javanmard *et al.* (2009), the better growth in monocrop than in intercrop may be related to better light incidence and higher nutritional area available. Independent of type of intercropping, it was observed higher (P<0.05) stem diameter in dwarf sorghum. However, similar results were observed for giant sorghum with low or high seeding rates, reaching values of 15.0 and 14.2 mm, respectively. Due to the relationship between height and stem diameter, only treatments with giant sorghum showed bending, being higher when intercropped with beans, which can be a result of its climbing characteristic, imposing more weight to the sorghum support. There was no effect of seeding rate on the leaf: stem ratio in dwarf sorghum treatments. However, giant sorghum intercropped in medium or high seeding rates had higher (P<0.05) leaf:stem ratios, due to the higher percentage of leaves associated with reduced height. Consequently, in these conditions, the giant sorghum showed reduced stem percentage, verifying values similar to the dwarf sorghum.

The association of dwarf sorghum with soybean increased sorghum grain/panicle production (Table 1), mainly when seeded at the medium rate. In contrast, bean intercropped with giant sorghum did not affect the production of grain/panicle compared with sorghum in monoculture. However, intercropping at low seeding rate had higher grain yield than medium and high seeding rates (188.9, 120.3 and 117.1 g kg^{-1} of FM, respectively). Dwarf sorghum intercropped with soybean had the same forage yield (DM) as in

Table 1. Morphological and yield aspects of sorghum intercropped with legumes at different seeding rates.

Treatments[1]	Sorghum plants/ha (thousand plants)	Height (m)	Stem diameter (mm)	Plant bending index	Leaf: stem ratio	Grain+ panicle (g kg^{-1} FM)	Sorghum yield (Mg DM ha^{-1})	Total yield (Mg DM ha^{-1})
DS	196.0^{ab}	1.65^{b}	14.12^{b}	0^{c}	0.56^{a}	194.8^{bc}	13.12^{abc}	13.12^{bc}
DSSL	144.9^{cd}	1.71^{b}	15.37^{ab}	0^{c}	0.68^{a}	232.72^{ab}	14.19^{ab}	15.98^{ab}
DSSS	170.5^{bc}	1.58^{b}	16.37^{a}	0^{c}	0.67^{a}	259.04^{a}	14.49^{a}	16.65^{a}
DSSH	159.1^{cd}	1.53^{b}	15.62^{ab}	0^{c}	0.73^{a}	222.15^{ab}	10.52^{cd}	13.54^{bc}
GS	210.2^{a}	2.04^{a}	11.25^{c}	0.59^{bc}	0.30^{b}	149.58^{cd}	13.58^{abc}	13.58^{bc}
GSBL	139.2^{d}	2.00^{a}	15.00^{ab}	1.35^{a}	0.29^{b}	188.9^{bc}	11.30^{bcd}	12.31^{c}
GSBS	150.6^{cd}	1.72^{b}	11.00^{c}	1.39^{a}	0.56^{a}	120.33^{d}	8.73^{de}	10.76^{c}
GSBH	144.9^{cd}	1.66^{b}	14.25^{b}	1.19^{ab}	0.56^{a}	117.14^{d}	5.68^{e}	7.31^{d}
Mean	164.4	1.7	14.1	0.6	0.5	185.6	11.5	12.9
SEM	9.21	0.07	0.70	0.23	0.06	18.56	1.08	1.04

[1] DS = dwarf sorghum; DSSL = dwarf sorghum plus soybean low seeding rate; DSSS = dwarf sorghum plus soybean medium seeding rate; DSSH = dwarf sorghum plus soybean high seeding rate; GS = giant sorghum; GSBL = giant sorghum plus bean low seeding rate; GSBS = giant sorghum plus bean medium seeding rate; GSBH = giant sorghum plus bean high seeding rate; SEM = standard error of the mean; FM = fresh matter; DM = dry matter.

monoculture. However, dwarf sorghum at low and medium seeding rates showed higher yields than when intercropped at high rate of soybean seeding (14.1, 14.4 and 10.5 t DM ha^{-1}, respectively), indicating a high competition among plants. For giant sorghum, the inclusion of higher seeding rate of bean decreased forage mass production, with smaller productions in treatments with high and medium seeding rates than in sorghum monoculture (5.7, 8.7 and 13.6 t DM ha^{-1}, respectively). Among all treatments, there was better compensation for the production of dwarf sorghum intercropped with soybean at medium seeding rate, which achieved a total forage mass productivity (sorghum plus legumes) of 16.6 t DM ha^{-1}, while the lowest compensation was found for association between giant sorghum and beans, at high seeding rate (7.3 t DM ha^{-1}).

Conclusions

Dwarf sorghum intercropping with soybeans at standard seeding rate (15 seeds m^{-2}), seems to be the best alternative to increase productivity and forage quality, without compromising morphological development of sorghum in the field. Thus, the practice of intercropping between grasses and legumes may be a good strategy to optimize the sustainable forage production systems, since observed the compatibility between intercropped species.

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Fatty acid, carotenoid and vitamin-E contents of *Plantago lanceolata* at different maturity stages

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Abstract

Ribwort plantain is among the most common broad-leaved unsown forb species found in Galician grasslands. A study was conducted to evaluate the changes in the vitamin (carotenoids and tocopherols) and fatty acids (FA) contents. In a pasture in the second year of production sown with a mixture ryegrass-red clover, samples of plantain were clipped at three weeks interval from May to June in three growth stages (vegetative, intermediate and mature). The main carotenoids in the dry matter (DM) were lutein and all-*trans*-β-Carotene, followed by violaxanthin and neoxanthin (averaged values of 140.1, 92.6, 44.8 and 30.8 mg kg^{-1} DM, respectively). Lutein and neoxanthin contents increased towards the end of spring, whilst α-tocopherol content was numerically higher in the vegetative stage. Digestibility, crude protein content and FA concentration decreased with maturity stage. In conclusion, the present study suggests that ribwort plantain can be a valuable pasture resource, at least in the early grazing season of the year.

Keywords: pasture, carotenoids, vitamins, digestibility, fatty acids

Introduction

Grasslands represent a cheap feed for ruminants and provide an important natural source of vitamins and fatty acids (FA). Their concentrations in forage species are important for the quality of animal derived food products (milk and meat). Pastures with a more diverse botanical composition, including forbs, may be an opportunity for obtaining animal products with higher quality and market value. Ribwort plantain (*Plantago lanceolata* L.) is among the most common broad-leaved unsown forb species found in Galician grasslands, being highly palatable for the grazing animals. Elgersma *et al.* (2013) reported that this forb enhances vitamin and FA concentrations beneficial for ruminant feeding. Until the date there is not local information available about the nutritive quality of ribwort plantain in grazing systems of Galician dairy farms. The objective of this work was to evaluate the evolution of chemical composition, digestibility, FA profile, carotenoids and vitamin-E contents of this specie during the spring of 2013.

Materials and methods

The present work was carried out at the Centro de Investigacións Agrarias de Mabegondo research station farm (Galicia, NW Spain: 43° 15′ N, 8° 18′ W, 100 m above the sea level) from 7 May to 18 June 2013. In a pasture in the second year of production, sown with a mixture of perennial ryegrass-red clover, three samples (0.36 m^2) were randomly collected, every three weeks, for each growth stage: vegetative (no emerged inflorescences), intermediate (flowering) and mature (end of flowering). The herbage samples were manually cut to a 5 cm stubble height and the whole plantain were separated and divided into two subsamples to determine: (1) dry mater content, nutritive value and FA concentration and (2) carotenoid and vitamin-E contents. All analytical determinations were performed in duplicate. Dry matter (DM) content was determined by oven-drying (60 °C, 48 hours) and dry samples were ground in a Christy-Norris hammer mill with a 1 mm screen. The chemical composition and digestibility were determined as described by Flores *et al.* (2003) by reference methods. The FA were extracted using a modified version of the direct transesterification method developed by Sukhija and Palmquist (1988), performing a simultaneous extraction-methylation step with methanolic hydrochloric acid. The FA methyl esters were detected and quantified by GC coupled with a flame ionization detector. The intact subsamples for carotenoid and vitamin-E determinations were immediately vacuum packed (Tecnotrip EV-15-1CD, Terrasa, Spain) and frozen (-20 °C) until posterior analysis (<1 month). Tissue disintegration (by cutting and blending in a food processor under liquid N_2) and residual moisture measurement were carried out immediately before extraction. Simultaneous extraction of carotenoids and vitamin-E was conducted according to Cardinault *et al.* (2008) and to the saponification procedure described by Chauveau-Duriot *et al.* (2010). Separation and quantification of compounds was carried out on an HPLC system coupled with a photodiode array (carotenoids) and a scanning fluorescence (vitamin-E, excitation-emission at 295-330nm) detectors under similar chromatographic conditions to those described by Chaveau-Duriot *et al.* (2010). Data were subjected to ANOVA and comparison of means by Duncan's Multiple Range test using Proc GLM of SAS (SAS Institute, 2009).

Results and discussion

On average, unsown ribwort plantain represented 20% of total pasture biomass. Its nutritive value can be considered as intermediate between that of perennial ryegrass and red clover, with a content in crude protein (CP) ranging from 122 to 94 g kg^{-1} DM, high *in vitro* digestibility (IVOMD) ranging from 830 to 694 g kg^{-1}, associated with a low cell-wall content (NDF) in the interval 349 to 432 g kg^{-1}DM. The plantain quality decreased with advancing maturity, and a reduction in the CP content of -3.5 g kg^{-1}

Table 1. Fatty acid, carotenoid and vitamin-E contents of ribwort plantain at three maturity stages.[1]

Stages of maturity	**V (n=3)**	**I (n=3)**	**M (n=3)**		
Date	**7 May**	**28 May**	**18 June**	**sem**	***P***
Main individual FA (g kg^{-1} DM)					
Palmitic	2.59	2.11	1.78	0.030	<0.0001
Stearic	0.29	0.20	0.19	0.004	<0.0001
Oleic	0.56	0.47	0.46	0.003	<0.0001
Linoleic	2.22	1.77	1.40	0.029	<0.0001
Alpha-linolenic	6.48	5.52	4.29	0.158	<0.0001
Summations of FA classes (g kg^{-1} DM)					
Total FA	12.41	10.32	8.35	<0.0001	0.166
Saturated FA	3.03	2.47	2.13	<0.0001	0.044
Monounsaturated FA	0.61	0.50	0.49	<0.0001	0.002
Polyunsaturated FA	8.78	7.35	5.73	<0.0001	0.157
Carotenoids (mg kg^{-1} DM)					
Neoxanthin	22.43	29.82	40.43	0.0056	3.313
Violaxanthin	40.63	43.56	50.47	0.4471	5.484
Antheraxanthin	1.45	4.06	1.61	<0.0001	0.202
Lutein	105.78	142.07	174.59	0.0065	12.852
Zeaxanthin	1.83	2.42	2.30	0.0338	0.151
β Cryptoxanthin	0.26	0.67	0.67	<0.0001	0.040
All-*trans*-β-Carotene	86.12	90.65	101.17	0.4315	8.188
13-*cis*-β-Carotene	2.95	2.37	3.49	0.2269	0.438
Vitamin-E (mg kg^{-1} DM)					
α-tocopherol	29.65	20.91	15.94	0.0595	3.587
γ-tocopherol	2.32	2.29	2.05	0.7870	0.275

[1] V = vegetative; I = intermediate; M = mature; DM = dry matter; FA = fatty acids; sem = standard error of the mean.

DM week^{-1} and a more marked decline in IVOMD (-17 g kg^{-1} week^{-1}) were observed. Warner *et al.* (2010) reported values of plantain NDF between 174 and 295 g kg^{-1} DM) and CP between 87 and 115 g kg^{-1} DM, and Søegaard *et al.* (2011) reported lower IVDMO, higher NDF and similar CP contents than the values obtained in the present work. The advance towards maturity significantly (P<0.0001) decreased individual and total FA concentration *in planta*in DM, but it did not alter the relative FA percentages, with average values for alpha-linolenic (ALA), palmitic (PA), linoleic (LA), oleic and stearic FA of 524, 208, 173, 48 and 22 g kg^{-1} of total FA (TFA), respectively. The range of the concentration (in g kg^{-1} DM) was 12.41 to 8.78 for TFA, 6.48 to 4.29 for ALA, 2.59 to 1.78 for PA and 2.22 to 1.40 for LA (Table 1). These values are lower than those reported by Elgersma *et al.* (2013) probably reflecting different genotypic and environmental conditions. The main carotenoids were lutein and all-*trans*-ß-Carotene, followed by violaxanthin and neoxanthin (mean values of 140.1, 92.6, 44.8 and 30.8 mg kg^{-1} DM, respectively). Lutein and neoxanthin contents increased (P<0.001) towards maturity, as well as all-*trans*-ß-Carotene, although this trend did not reach significance (P>0.05). A negative relationship trend (P=0.059) between the plantain α-tocopherol content and age was detected, as was observed elsewhere (e.g. Ballet *et al.*, 2000) in other forage species. It is noteworthy to indicate that plantain α-tocopherol values are higher than those observed for perennial ryegrass and red clover (values not shown in this paper), whilst those of lutein and all-*trans*-ß-Carotene are intermediate.

Conclusions

The results of the present work suggest that ribwort plantain can be a valuable pasture resource in early spring, in terms of nutritive value and as a source of PUFA, carotenoids and vitamin-E in the ruminant diets.

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Statistical models to estimate the potential forage quality of permanent meadows at the first cut

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Abstract

Forage quality is known to change with the phenological development of plants. A large data set, encompassing a wide range of climatic conditions and management practices was obtained in permanent meadows in South Tyrol (NE Italy) by means of sequential sampling and analysis of fresh-cut forage for a period of seven weeks starting at the phenological stage of stem elongation. Sampling was performed at 202 environments from 2003 to 2014 at altitudes between 666 and 1,593 m a.s.l. Statistical predictive models, taking into consideration meteorological and climatic variables (primarily growing degree days), as wells as variables related to geomorphology, botanical composition, soil and agronomic management were developed for crude protein (CP) and K by means of mixed models. They were optimised using a stepwise forward selection and subsequent five-fold cross-validation. Four models per each parameter, based on different combinations of available predictor variables, were developed. Higher predictive accuracy was found for the models taking the entire set of independent variables into account. The best predicted quality parameter was CP, while lower prediction accuracy was found for K. Lack of correlation was the most relevant component of the mean squared deviation of the models.

Keywords: permanent meadows, forage quality, growing degree-days, statistical predictive models, model evaluation

Introduction

Forage quality can best be ascertained by means of laboratory analyses, but reference values can be obtained from tabulated forage values (German: Futterwerttabellen), which are mainly based on the relationships between the quality of forage and the phenological development of plants (Resch *et al.*, 2006). A first attempt at estimating forage quality depending on variables related to climate and meteorology, as well as to the botanical composition, geomorphological factors, soil properties and the management of permanent meadows was successfully performed by means of binary logistical regression using a large dataset obtained during a long term study on the forage quality at the first cut in South Tyrol (Peratoner *et al.*, 2010). In this paper, the development of statistical predictive models, based on this dataset, in order to achieve a quantitative prediction, is exemplarily illustrated for crude protein (CP) and K.

Materials and methods

Forage samples were weekly sequentially sampled in permanent meadows in fourfold replications for a period of seven weeks starting at the phenological stage of stem elongation (15 cm growing height) at 202 environments (site × year combinations) from 2003 to 2014 at altitudes between 666 and 1593 m a.s.l. in South Tyrol (Italy). All parameters but *in vitro* digestibility and minerals were analysed by means of NIRS (NIRSystem 5000, FOSS, U.S.A.). 20% of the samples collected between 2003 and 2007 were analysed by wet chemistry according to Naumann *et al.* (1997) in order to establish a suitable calibration curve. Mineral elements were determined according to Naumann *et al.* (1997) in mixed samples obtained by pooling the four replicates obtained at each sampling event.

Sampling and survey of geomorphologic factors, soil properties and information related to management practice and botanical composition were as described in Peratoner *et al.* (2010) and Peratoner *et al.* (2016a). The following meteorological and climatic variables were also used for the development of statistical predictive models: (1) growing degree-days (GDD) based on observed temperatures (2 m above the ground) from representative weather stations for 20 of the 35 experimental sites and on interpolated temperatures (250×250 m spatial resolution) according to Schaumberger (2011) for the other sites. Base temperatures according to Romano *et al.* (2014) were used to compute GDD; (2) sum of the potential global radiation sums calculated on topographic base with the Solar Analyst tool of ArcGIS at a resolution of 100 m; (3) the sum and the average of daily departures of rainfall from a long-term daily average (20 years) of reference weather stations within homogeneous precipitation districts. Precipitation districts were defined by means of a cluster analysis (monthly precipitation sums between 2001 and 2013 as variables, squared Euclidean distance as dissimilarity measure, Ward's method as clustering algorithm), grouping the weather stations of the Province of Bozen into five homogeneous groups and assigning the respective catchment areas as well as the surrounding ones to the respective group. For each forage sample 1), 2) and 3) were calculated for the time interval between one week before the date of stem elongation and the cutting time. The statistical predictive models were developed by means of mixed models starting from a baseline model accounting for the design effects and the effect of GDD (Romano *et al.*, 2014), which was optimised using a stepwise forward selection, until no further improvement of the Akaike Information Criterion (AIC) was achieved. Effects with $P>0.1$ were dropped from the model, with the exception of marginal effects of polynomials or main effects for interactions (Nelder, 2000). Subsequently, the model was further developed by stepwise forward selection and backward elimination, until the squared correlation between observed and predicted values in five-fold cross-validation (Hawkins *et al.*, 2003) peaked. The values predicted by the fixed part of the model were used. Partitioning of the mean squared deviation according to Gauch *et al.* (2003) was used to evaluate the models. Data transformation was performed as necessary to achieve normal distribution and homoscedasticity of residuals. For each quality parameter, four models were developed: using all variables (ALL), without using the meadow type (OS), without using the soil properties (OV) and without using both the meadow type and the soil properties (OBL).

Results and discussion

The statistical models for CP and K were found to differ concerning the prediction accuracy, which was higher for CP than for K (Table 1). Increasing the number of variables taken into account for model development increased for both CP and K the predictive accuracy. This increase was small for CP (from 0.680 to 0.702) and quite relevant for K (from 0.434 to 0.563). For both quality parameters, the

Table 1. Prediction accuracy and partitioning of the mean squared deviation of predictive models for CP and K. MSD = mean squared deviation, SB = squared bias, NU = nonunity slope, LC =lack of correlation. ALL: model developed with all available variables, OS: without the meadow type, OV: without the soil properties, OBL: without soil properties and meadow type.

Model	Quality parameter CP[1]					K				
	R^2	MSD	SB	NU	LC	R^2	MSD	SB	NU	LC
ALL	0.702	1.119	0.002	0.001	1.116	0.563	0.354	<0.001	<0.001	0.353
OS	0.686	1.181	0.002	0.001	1.178	0.547	0.372	<0.001	<0.001	0.373
OV	0.680	1.203	0.003	0.002	1.198	0.479	0.393	<0.001	<0.001	0.393
OBL	0.680	1.203	0.003	0.002	1.198	0.434	0.403	<0.001	<0.001	0.403

[1] Analysis with square root-transformed data.

increase of prediction accuracy achieved by taking into account the soil properties (OS vs OBL) was higher than the increase obtained by using the meadow type (OV vs OBL). The additive components of the mean squared deviation (MSD) of the models according to Gauch *et al.* (2003) showed low values for squared bias (SB) and nonunity slope (NU). SB quantifies the translation of the intercept of the regression line between predicted and observed values from 0, whilst NU quantifies its rotation, intended as the departure of the slope from unity. Lack of correlation (LC) was the most relevant component. The models developed taking into account all variables had the lowest LC.

Conclusions

Statistical models estimated CP with reasonable precision, while lower values were attained for K. The use of all variables resulted in the highest prediction accuracy, but the magnitude of the improvement depended on the investigated parameter. Lack of correlation was the main error source. These models, along with all other models developed with the same method for further quality parameters, have been implemented into a web-application for practical use in grassland farming (Peratoner *et al.*, 2016b).

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Effects of yeast inoculation and air exposure on the nutritive value of corn silages

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Abstract

Corn silages are prone to deterioration when exposed to air. The lactate-assimilating yeast species are frequently the first microorganisms to initiate the aerobic deterioration. Some studies reported that aerobically unstable corn silage is associated with reduced nutritive value. Therefore, the objective of this study was to evaluate the influence of yeast inoculation (*Pichia norvegensis*) and the air exposure by 48 hours on the fermentative profile and digestibility of corn silages. Corn crop was harvested at 34% DM, treated without (Control) or with *P. norvergensis* at 1×10^5 cfu g^{-1} fresh matter (Yeast) and packed in large scale bag silos (40 t $silo^{-1}$). After 123 d of storage, silos were opened. Daily, silages were unloaded and fed to cows immediately (Fresh) or after 48 h of air exposure (Exposure). The four treatments were: control-fresh silage (CF), control-exposed silage (CE), yeast inoculated-fresh silage (YF), and yeast inoculated-exposed silage (YE), in a factorial design. The effects of air exposure by 48 hours reduced lactic acid concentration ($P<0.001$), increased the pH ($P=0.004$) and also changed other acids, alcohols and esters concentrations. Moreover, there was a trend of decrease for the digestibilities of DM, NDF and TDN.

Keywords: aerobic deterioration, yeast, fermentative profile, digestibility

Introduction

Corn silages are very prone to deterioration, due to the forage temperature rise as consequence of respiration of aerobic microorganisms. Moreover, according to the literature, the microorganisms which start the deterioration when silages are exposed to air in the feedout phase are the lactate assimilating yeasts and the most common genera related are *Pichia* and *Candida*. Some studies have shown that silages exposed to air and with high counts of yeasts, decreased the nutrient digestibility and altered the fermentative profile, followed by a decrease in intake, milk yield and milk fat content. Thus, the objective of this study was to evaluate the effect of *Pichia norvegensis* inoculation and air exposure by 48 hours on the nutritive value and fermentative profile of corn silages.

Materials and methods

Corn silage was harvested with 34% of dry matter at Piracicaba, Brazil. The silage was stored in large scale bag silos (40 ton/silo) and after 123 days the silos were opened. In a previous trial, carried out in Brazil, it was identified that the species *Pichia norvegensis* was the most frequent yeast in corn silage samples, using a commercial yeast identification kit (ID 32C, bioMerieux, Marcy l'Etoile, France). In this way, at the ensiling, *P. norvegensis*, diluted in a molasses solution, was inoculated at the concentration of 1×10^5 cfu g^{-1}, creating an yeast inoculated silage (Y) and similar amount of molasses was applied to control silage (C), in order to equalize the sugar concentration on both treatments. Treated silages were offered to Holstein cows for performance evaluation (data not shown) for four periods and the samples were collected at d-15 of the period (21d). In each period, samples of the both silages were collected immediately after silo unloading (F) and after 48 hours of air exposure (E). In this way, the treatments were set by considering a factorial design: control-fresh silage (CF), control-exposed silage (CE), yeast inoculated-fresh silage (YF), and yeast inoculated-exposed silage (YE). To estimate the nutritive value of the corn silages, samples were collected to measure the pH, fermentative profile and nutrients digestibilities. The pH was measured according to the methodology described by Kung Jr. *et* *al.* (1984) and the lactic acid content was measured by using a spectrophotometer as described by Pryce (1969). Samples were centrifuged (10,000×*g*) and filtered to indentify and quantify the fermentative profile using the method of gas chromatography with mass spectrometry. The *in vivo* digestibility was estimated considering the indigestible NDF after 288 hours of rumen incubation as a marker (Huhtanen *et al.*, 1994), to estimate faecal production of the Holstein cows.

Results and discussion

The characteristics of corn silage fermentation and its DM, NDF and total nutrients digestibilities (TDN) are described in Table 1. According to the data, the inoculation of *Pichia norvegensis* did not affect negatively the corn silage, as much as the air exposure for 48 hours did. However, the inoculation with the chosen yeast strain only caused a trend of decrease of the ethanol content and it could be supplied by the capacity of yeasts to assimilate alcohols and other compounds in aerobic environments. On the other hand, when corn silage was exposed to air, the negative changes were very clear, showing a trade-off effort in between decrease of desirable (lactic and acetic acids, which are responsible to drop the pH and therefore conserve the roughage under anaerobic condition) and increase of undesirable compounds (propionic, formic, isobutyric acids and 2,3-butanediol that are result of pathogenic microorganisms metabolism). The pH was increased when silages were exposed to air, possibly as a result of the decrease in lactic acid and acetic acid contents of these silages. The quantity of ethanol and ethyl lactate, also decreased with the air exposure. During the feedout (aerobic phase) some microorganisms are responsible to initiate the aerobic deterioration of silages and they are specific for lactate consumption and the most known microorganisms are the lactate-assimilating yeast species. Although the yeasts can metabolize the lactate, it also has a variable capacity of utilizing ethanol for development (Pahlow *et al.*, 2003). Moreover, according to Spoelstra *et al.* (1988), acetic acid bacteria are capable of oxidizing organic compounds, such as acetic acid, and also initiate the silage spoilage. Furthermore, after beginning the aerobic deterioration, harmful microorganisms can develop and reduce the hygiene quality of the silage. The concentrations of propionic, formic, isobutyric acids and 2,3-butanediol increased in the silages exposed to air, though there was no change in butyric acid concentration. All the compounds described before are possible to

Table 1. Fermentative profile and nutrients digestibility of corn silages inoculated with *Pichia norvegensis* and/or exposed to air by 48 hours.

	Control		Yeast			*P*-value[1]		
Item	Fresh	Exposed	Fresh	Exposed	SEM	Y	E	Y×E
pH	3.79	4.35	3.55	4.29	0.17	0.27	0.004	0.48
Lactic acid, % DM	4.33	1.71	4.67	2.05	0.51	0.52	0.001	0.99
Acetic acid, % DM	1.82	1.59	1.99	1.38	0.28	0.92	0.05	0.33
Ethanol, % DM	1.12	0.22	0.35	0.05	0.26	0.06	0.03	0.22
Propionic acid, % DM	0.04	0.23	0.04	0.20	0.07	0.75	0.01	0.85
2,3-Butanediol, % DM	0.04	0.52	0.03	0.55	0.15	0.92	0.003	0.88
Ethyl lactate, mg kg^{-1}	148	10	117	14	16.27	0.36	<0.001	0.23
Formic acid, mg kg^{-1}	126	1811	89	1750	601	0.93	0.01	0.98
Butyric acid, mg kg^{-1}	122	84	124	119	33	0.52	0.44	0.56
Isobutyric acid, mg kg^{-1}	32	197	41	215	35	0.71	0.001	0.90
DMD, %	71.57	70.49	71.16	69.97	0.67	0.45	0.07	0.93
NDFD, %	55.07	53.17	54.41	53.90	0.83	0.96	0.08	0.31
TDN, %	70.73	69.81	70.11	69.13	0.60	0.25	0.09	0.96

[1] Effects of inoculation with *P. norvegensis* (Y), air exposure by 48 hours (E) and the interaction (Y×E).

be from *Clostridium* metabolism, even though identification of this species was not performed. The high increase of formic acid is unknown due a lack of information about the quantity of microorganisms present in silage during aerobic exposure. Regarding the nutrients digestibility of the silage exposed to air, there was a trend of decrease of DMD, NDFD and TDN. The major contribution in decrease the silage digestibility came from the reduction of NDFD. According to Coblentz and Hoffman (2009), the spontaneous heating of the forage might reduce the digestibility of NDF by increasing the presence of hemicellulose in Maillard and other reactions.

Conclusions

Exposing corn silages to air by 48 hours, as expected, can reduce the concentration of compounds that are important in forage conservation, such as lactic and acetic acids, and also increase the compounds that are undesirable (propionic, formic, isobutyric acids and 2,3-butanediol), which represent the growth of harmful microorganisms in silage. Moreover, the air exposure can cause a trend of decrease on the nutrients digestibilities.

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Suitability of different methods to describe the botanical composition for predicting forage quality of permanent meadows at the first cut

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Abstract

The botanical composition is relevant to the forage quality of permanent meadows, as species differ from each other in their quality characteristics. A complete and detailed description of the botanical composition and the classification into a meadow type (MT) is time-consuming and requires advanced botanical skills. A quicker and easier method that is often used for the estimation of the forage quality is to assign the plant stand to one of the plant stand types (PT) 'rich in grasses', 'balanced', 'rich in forbs' or 'rich in legumes' based on the yield proportion of grasses, forbs and legumes. A large data set of about 6,000 forage samples obtained by sequential sampling at 202 environments in South Tyrol (Italy), describing the changes in forage quality along the phenological development starting from stem elongation, was used to investigate the effect of both MT and PT on 16 parameters of forage quality. Starting from a baseline model including design effects and growing degree days as a covariate, the improvement of the prediction accuracy of the model due to MT and PT was described by a five-fold cross-validation of stepwise forward-developed mixed models. PT contributed to improve the prediction accuracy of most of the investigated quality parameters and for 10 parameters was taken first into the model. MT was relevant as well for some of the parameters, but it played a less consistent role.

Keywords: forage quality, botanical composition, plant stand type, meadow type, permanent meadows

Introduction

It is well known that forage quality and its phenology-related changes over time are species-specific (Jeangros *et al.*, 2001; Bruinenberg *et al.*, 2002). For this reason, the botanical composition of multi-species plant stands is often taken into account for the prediction of the expected forage quality. Because of the effort needed to fully describe the botanical composition of the plant stand, a simplified system only considering the proportion of the species groups, such as grasses, legumes and forbs is used (Daccord *et al.*, 2007). The present paper addresses the question, whether a detailed description of the botanical composition, leading to the classification of the plant stand into a vegetation type, may improve the predictive accuracy of the parameters of forage quality.

Materials and methods

In order to answer the experimental question, a large data set was used, which describes the changes over time of the forage quality of permanent meadows between 666 and 1,593 m a.s.l. in South Tyrol (Italy) at the first cut. Forage samples were taken in fourfold replication per sampling event between 2003 and 2014 at 202 environments (site × year) by means of weekly sequential sampling for seven weeks starting at the stage of stem elongation (15 cm herbage growing height) and analysed for quality parameters (Peratoner *et al.*, 2016). Details about forage analyses are given in Romano *et al.* (2016). Growing degree-days (GDD) were computed for each sample according to Romano *et al.* (2014). For minerals and *in vitro* digestibility the four replications were pooled to one sample. Prior to harvest of each forage sample, the yield proportion of grasses, legumes and forbs was visually estimated and each sample was assigned to a plant stand type (G = rich in grasses: more than 70% grasses; B = balanced: between 50% and 70% grasses; F = rich in forbs: more than 50% forbs, legumes less than 50%; L = rich in legumes: legumes more than 50%) according to Daccord *et al.* (2007). In contrast to the original method, no sub-classification of G and B based on the proportion of ryegrass-species and of F based on the proportion of forbs rich in stems was made. The plant stand type of mixed samples was determined according to Table 1.

Once per year, at the time of the third or fourth sampling event, the yield proportion of each occurring species was assessed at each environment. Each site was assigned to one of seven meadow types according to a cluster analysis of these data, which were averaged across the years and transformed according to Dietl (1995), using the squared Euclidean distance as dissimilarity measure and the Ward's method as a clustering algorithm. The statistical analysis was performed by means of mixed models (proc MIXED of SAS version 9.2), accounting for the design effects and for the effect of GDD, which was modelled by means of a polynomial regression (Romano *et al.*, 2014). The improvement of the prediction accuracy due to the inclusion of PT and MT into the model was investigated by means of a stepwise forward selection based on a five-fold cross-validation according to Hawkins *et al.* (2003). The predicted values of the fixed effects of the model were used. If necessary, data transformation was performed to achieve normality and homoscedasticity of residuals.

Table 1. Classification scheme of mixed samples obtained by pooling the four replications harvested at each sampling event.[1]

Plant stand type of the four replications	Classification of the mixed sample
At least 3 G	G
2 G and 2 B	G
At least 3 F	F
2 B, all other samples F or L	F
All other combinations	B

[1] G = rich in grasses; B = balanced; F = rich in forbs; L = rich in legumes.

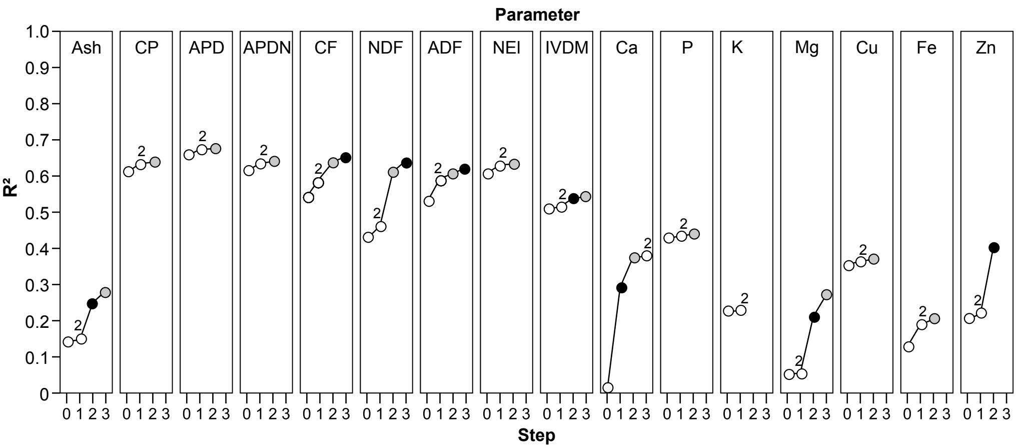

Figure 1. Changes of the prediction accuracy (squared correlation between predicted and observed value of the five-fold cross-validation) by stepwise forward model selection for 16 parameters of forage quality. White dots= GDD (2 indicates the quadratic term of the polynomial); grey dots = PT; black dots = MT.

Results and discussion

The prediction accuracy for ash and minerals was apparently lower than for all other parameters (Figure 1). The relationship between GDD and all quality parameters was best described through a quadratic polynomial. PT was added to the model in 14 of the 16 cases, with the exception of K and Zn, whilst MT contributed only for half of the quality parameters to improve the prediction accuracy. PT systematically exhibited higher relevance than MT for the prediction accuracy of crude protein (CP), absorbable intestinal protein (APD), absorbable intestinal protein based on the nitrogen available in the rumen (APDN), crude fiber (CF), neutral detergent fiber (NDF), acid detergent fiber (ADF) and net energy for lactation (NEl). For ash, *in vitro* digestibility (IVDM) and minerals PT exhibited a minor relevance. Only for four parameters (ash, Ca, Mg and Zn) the addition of MT to the model resulted in a notable improvement of the prediction accuracy.

Conclusions

A simple assessment of the proportion of grasses, forbs and legumes and the classification of the plant stand into a category, which represents a reasonable effort for practitioners, is a useful tool for improving the prediction of forage quality. More precise assessments of the botanical composition, resulting in the definition of a meadow type, improve the prediction accuracy of some parameters, but play a less consistent role.

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Breeding of cocksfoot (*Dactylis glomerata* L.) with improved forage quality

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Abstract

Cocksfoot (*Dactylis glomerata* L.) is a valuable species of cool-season perennial grasses that can be used for various purposes. It has many advantages: high productivity, rapid development, drought resistance, persistence and good winter hardiness, which is particularly important for the northern latitudes. The greatest shortcoming of cocksfoot is its low palatability due to the harshness of leaves having silicified dentations. Therefore, it has been a challenge for the cocksfoot breeders to create a high productive variety with soft leaves i.e. good palatability. Newly developed cocksfoot breeding lines from 2012 to 2015 were evaluated for dry matter yield (DMY) and forage quality in comparison with the most productive and commonly used varieties in Latvia. Regardless of the fact that in general there is a negative correlation between DMY and palatability of cocksfoot, some promising breeding lines which combine desirable properties have been created and selected. They might be used as a source material for new variety. In the four-year period the breeding line Jum/573 distinguished by softer leaves and significantly higher average DMY level.

Keywords: *Dactylis glomerata*, breeding, yield, forage quality

Introduction

Its very early and rapid development in the spring, good winter hardiness, persistence, drought tolerance (Santen and Sleper, 1996) and suitability for the cultivation in sandy soils (Fraser, 1994), make cocksfoot (*Dactylis glomerata* L) a very valuable species. At high rates of nitrogen, it is one of the most productive cool-season grasses (Lacefield *et al.*, 2003). However, efforts should be done for improving the digestibility (Dijk, 1959) and foremost palatability to mitigate the harshness of leaves due to the presence of silicified dentations. Therefore, breeders are continuously working on the creation of a high productive variety with soft leaves or better palatability. Some prospective breeding lines have been created as a result of many years activities (starting from the late 1980s), including the repeated crossing of desirable forms and prolonged individual and family selection. The aim of our studies was to select among them the most productive and persistent cocksfoot breeding lines with soft leaves and good forage quality. We summarize here data of five promising cocksfoot breeding lines and compare them with three widely used cocksfoot varieties in Latvia, including the only local variety 'Priekulu 30', which is high productive, winter-hardy and persistent. However, it has very harsh leaves and hence a low palatability. Therefore, work is continuously going on to create a new high productive variety with «soft leaves».

Materials and methods

The breeding work of cocksfoot is carried out in the central part of Latvia (56°37 N and 25°07 E). In order to assess the breeding material 18 cultivars and breeding lines of cocksfoot were included in a randomized block design with three replicates, seeding rate of 15 kg ha^{-1}. The trial was established in July of 2011 without cover crop in a sod-podzolic loam soil, pH KCl 5.3, plant available P_2O_5 167 mg kg^{-1}, K_2O 88.0 mg kg^{-1}, OM content 27 g kg^{-1}. We report here data of 4 consecutive ley years (2012-2015) of five more promising breeding lines and three commonly used varieties in Latvia. Before establishment of trial and after last harvest in the ley years compound fertilizers were used: N 15 kg ha^{-1}; P_2O_5 30 kg ha^{-1}; K_2O 75 kg ha^{-1}. In addition, N 60 kg ha^{-1} three times per ley year: just after the beginning of vegetation and after the 1st and 2nd cut was used. Winter hardiness was evaluated in spring every year just after the beginning of vegetation by assessing percentage ground cover. Always just before cutting the following was evaluated: softness of leaves by touch; colour of sward; resistance to diseases. The rating was expressed in points: 1 very harsh/9 very soft leaves; 1 very light green/9 very dark green colour of sward; 1 very weak/9 very strong resistance to diseases. All swards were harvested three times per growing season: 1st cut at the ear emergence (end of May or early June); the 2nd and the 3rd cut at approximately 40-50 days after previous cutting. Crude protein (Kjeldal method) and acid detergent fibre (ADF, gravimetrical method) were determined and dry matter digestibility (DDM, %) calculated from ADF: DDM, % = 88.9 – (0.779 × ADF, %). The experimental data were processed by analysis of variance (ANOVA), the differences among means were detected by LSD at the probability level of 0.05.

Results and discussion

The comparison between cocksfoot varieties and promising breeding lines showed that the breeding work and long-term selection contributed to select high productive breeding lines with relatively soft leaves. In general, cocksfoot has a relatively good winterhardiness in Latvian condition. A special situation arose in the winter of 2013/2014 when for about one month in January – February there was a strong blackfrost with cold wind, killing many crops. The assesment of cocksfoot in spring 2014 showed that some swards were partly damaged, but most of them recovered during the season suggesting that a succesful selection had been carried out. Trials data and experience of many years suggest that in terms of wintering the most stable varieties are 'Priekulu 30' and breeding line Jum/573 (Table 1).

Several breeding lines were distinguished for soft leaves, which substantially outperformed standard 'Priekulu 30'. However, the selection work in this area should be continued. On average, the protein content was satisfactory for all selected breeding lines. It was slightly lower for Jum/573 as it is the earliest of all varieties and lines included in the trial. In the vegetative (leafy) stage, cocksfoot is a high-quality forage. Quality declines as the plants approach maturity (Lacefield *et al.*, 2003).

The DMY of cocksfoot varieties and breeding lines during the four year period ranged from 7.02 Mg ha^{-1} to 8.96 Mg ha^{-1} on average (Table 2). In the first three ley years, yields of all varieties and breeding lines were quite stable, they ranged around the same level as in the first year of use – from 7.04 to 9.23 Mg ha^{-1}. The yield declined during the 4th year (2015): it varied between 5.77 and 7.87 Mg ha^{-1}. The breeding line Jum/573, which has relatively soft leaves, significantly (P<0.05) outyielded all other varieties. Usually cocksfoot varieties with softer leaves lag behind in terms of productivity.

Table 1. Evaluation of cocksfoot varieties and breeding lines during four ley years (2012-2015) in Latvia. Data are averages over cuts and years.

Variety/N°	Ground cover in 2014, %	Sward assesment in points (1-10)			Forage quality, % of DM		
		softness of leaves	colour	resistance to diseases	Crude protein, %	ADF, %	DDM, %
Priekulu 30	79.5	4.0	6.4	6.4	14.5	34.6	62.0
Amba	78.7	4.7	6.5	6.5	14.8	34.4	62.2
Aukstuole	77.5	5.5	8.0	7.5	14.7	34.9	61.7
D/570	75.0	5.7	5.0	7.5	15.5	33.0	63.2
Con/519	80.0	6.0	5.0	6.5	14.6	34.8	61.8
444/518	75.0	6.3	7.0	7.0	15.6	34.7	61.9
Jum/573	80.0	6.0	5.0	6.5	13.4	33.9	62.5
11/12/SL	80.0	6.0	5.0	7.5	14.2	34.8	61.8
$LSD_{0.05}$	2.7	1.1	1.5	0.6	1.8	1.7	1.3

Table 2. Dry matter yield (DMY) of cocksfoot varieties and breeding numbers in Latvia. Data are average of four ley years and 3 cuts per year.

Variety/N°	DM yield in three cuts, Mg ha^{-1}				
	2012	2013	2014	2015	Average
Priekulu 30	8.24	8.08	8.17	7.13	7.91
Amba	7.17	8.18	6.90	6.49	7.19
Aukstuole	7.04	8.45	8.37	5.77	7.41
D/570	7.05	7.04	8.9	6.84	7.46
Con/519	7.32	7.95	8.64	5.79	7.43
444/518	7.40	7.00	7.74	5.93	7.02
Jum/573	9.23	8.86	9.88	7.87	8.96
11/12/SL	8.64	7.87	8.39	6.65	7.89
$LSD_{0.05}$	1.08	0.65	0.52	0.85	1.04

DMY in all three cuts was similarly distributed over the season. Indeed, unlike many other grasses cocksfoot has a good regrowth capacity, the total yield is equally distributed over the year (Dijk, 1959). This evenly distributed DMY is one of the advantages of cocksfoot alongside with drought and heat tolerance (Santen and Sleper, 1996). With proper management and split applications of nitrogen, the part of the annual yield harvested after the first cut may contribute more than 50% of the total annual yield, as was confirmed in our trial. It provides an opportunity to gather fodder throughout the season.

Conclusions

As a result of prolonged breeding activities, some high productive promising breeding lines of *D. glomerata* with softer leaves and good winter hardiness were developed. They may be used in the further breeding work as a source material to develop new varieties. Breeding line Jum/573 combined softer leaves with a high DM yield in trial during four consecutive years.

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Yield and composition changes of temporary and permanent pasture

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Abstract

The changes in the botanical and chemical composition and enzymatic digestibility of one temporary (sainfoin and cocksfoot) and one permanent pastures are compared in two vegetative stages – without generative stems and in the emergence of generative stems for a period of three years. It was found that the dynamics of composition and digestibility are more intense in the temporary pasture compared to those in the permanent pasture. The yield of dry matter for the period between the stage without generative stems until the emergence of generative stems increases 2-3 times and is higher in the temporary pasture compared to that of the permanent pasture. The proportion of legume components and content of CP is higher in the temporary pasture in the first year and decreases over the next two years, the proportion of legumes is aligned with that of the permanent pasture. The enzymatic digestibility is higher in the stage without generative stems and decreases by 5 to 21% in the stages with generative stems in the temporary pasture and by 5 to 8% in the permanent pasture.

Keywords: temporary pasture, permanent pasture, botanical composition, chemical composition, digestibility, yield

Introduction

Permanent pastures are the most common sources for grazing and occupy the largest share in the feeding of sheep and goats in Bulgaria during the period from April to October. They represent about 30% of the total area for agricultural use, but this share decreases and the yield is low, on average 1,600 to 1,800 kg ha^{-1} hay (Kirilov and Mihovski, 2014). A good solution to this is to create temporary pastures with suitable composition of grasses and leguminous plants. The aim of this study is to compare the changes in the yield, botanical and chemical composition and enzymatic digestibility of a temporary and a permanent pastures in the stage without generative stems and in that with generative stems.

Materials and methods

The object of the study are one temporary and one permanent pastures located in the experimental field of the Institute of Forage Crops, Pleven, Bulgaria. The temporary pasture was planted in 2012 by cocksfoot (*Dactylis glomerata*) (25 kg ha^{-1}) and sainfoin (*Onobrychis viciifolia*) (120 kg ha^{-1}) in a ratio of 50:50 by sowing norm. Since the beginning of vegetation in April 2013, 2014 and 2015 plant samples in first undergrowth were taken of the two types of pastures for a period of four weeks. Every week an area of 1 m^2 was cut in four replications (n=4) and the biomass was weighed to determine the yield. The dry matter content, the share of grass and legume components are evaluated. The crude protein (CP) content, crude fiber (CF) (AOAC, 2007), Neutral-detergent fiber (NDF) and Acid-detergent fiber (ADF) are determined in dry samples according to Goering and Van Soest (1970), and the enzyme *in vitro* digestibility of dry matter (IVDMD) – according to Aufrere (1982). The results obtained during the first and second week of plant vegetation are united and consistent with the stage without generative stems, and those of the third and fourth weeks of the stage with generative stems. The data from the experiments were analysed by reporting the average (x) and standard deviation (± Sx) in the MS Office program Excel 2007.

Results and discussion

Dry matter yield is higher in the sown pasture compared to that of the permanent pasture (Table 1). In the first year of the experiment the yield from the sown pasture in the stage without generative stems (1-2 week) and generative stems (3-4 week) was up to three times higher than that in the permanent pasture. The same tendency of a three times higher yield in the sown pasture is observed over the next two years. The rate of increase in the yield in the temporary pasture is higher compared to that in the permanent pasture. In the temporary pasture during the stage of generative stems, the yield has increased 3 times compared to that in the stage without generative stems throughout all three years. A similar growth rate, but a lower one, is observed in the permanent pasture. In the first year (2013) after sowing, the share of legume component in the temporary pasture is equal to or higher than that of the grass and decreases in the second and third years. This reduction in the legume component is probably due to the low longevity of sainfoin (Frame, 2005). The proportion of legumes in the permanent grass covers ranges from 2.5 to 19.7%, no significant differences between the proportion of legumes in the temporary and permanent pastures are observed in the second and third years of the experiment.

In the stage without generative stems, the content of CP in both pastures is high and decreases during the stage with generative stems, and that of CF, NDF and ADF increases (Table 2). The decrease of CP in the temporary pasture is 19.8% in the first year, 26.1% in the second year and 43.5% in the third year, while in the permanent pasture the decrease is less expressed, respectively, 9.5, 21.5 and 25.8%. At the stage of generative stems in the temporary pasture during the first year NDF increased by 24.1% and in the second and third year respectively by 9.2 and 27.1%, for the same period the increase of NDF in the permanent pasture is respectively 11.6, 21.5 and 21.2%. A tendency towards lower levels of CP and higher such as structural carbohydrates is observed in the temporary pasture compared to the permanent pasture, which explains the respectively higher IVDMD in the permanent pasture. Digestibility of dry matter in the temporary pasture decreased by 5, 15 and 21% during the stage of generative stems, in three years respectively, and the decrease in the digestibility in the permanent pasture ranges between 5 and 8%. The differences in the values of the chemical composition and IVDMD are likely due to the rapid rate of growth and development of the temporary pasture where the yield during the three years of the experiment is higher than that in the permanent pasture.

Table 1. Dry matter yield (kg ha^{-1}), DM (g kg^{-1}) and botanical composition (grass:legume:other) of temporary (TP) and permanent pasture (PP) average (n=8) in stage without generative stems (1-2 week) and generative stems (3-4 week).

Parameter	2013		2014		2015	
	1-2 week	3-4 week	1-2 week	3-4 week	1-2 week	3-4 week
TP yield ($x \pm Sx$)	1,567.00±98.99	5,037.00±2,040.71	2,543.45±1,533.22	6,950.85±1,760.25.5	1,852.03±389.14	5,760.47±1,619.06
DM($x \pm Sx$)	174.55±4.31	186.0±4.67	212.90±18.67	257.35±6.86	187.00±2.83	218.56±4.95
gr:leg:other (1)	36.8:60.2:3	47:47:6	78.5:15.5:6	77:16.5:6.5	67.5:20.5:12	75.5:14.8:9.7
PP yield ($x \pm Sx$)	980.43±27.82	1,276.30±98.00	792.45±415.28	2,088.40±586.76	830.29±150.35	1,478.96±140.17
DM($x \pm Sx$)	182.95±7.00	208.90±15.70	218.70±30.41	288.85±7.99	210.00±18.38	255.00±14.14
grass:legume:other	93:2.5:4.5	85:8.5:6.5	75.6:14.4:10	72.8:19.7:7.5	77.7:10.9:11.4	77.3:8.9:13.8

Table 2. Chemical composition and IVDMD (g kg^{-1} DM) average (n=8) in stage without generative stems (1-2 week) and generative stems (3-4 week).

		Temporary pasture (x ± Sx)					Permanent pasture (x ± Sx)				
Vegetation week/year		CP	CF	NDF	ADF	IVDMD	CP	CF	NDF	ADF	IVDMD
2013	1-2 week	270.70	122.00	370.05	219.35	758.65	211.40	198.55	399.95	268.40	712.75
		±22.62	±16.82	±56.21	±2.33	±2.05	±1.13	±3.18	±72.8	±5.52	±14.78
	3-4 week	217.15	228.05	459.15	301.45	721.90	191.35	223.50	446.40	308.40	674.85
		±16.33	±9.26	±44.05	±27.79	±17.25	±0.64	±2.69	±22.2	±3.96	±13.08
2014	1-2 week	206.00	217.15	580.55	242.60	745.20	218.95	163.	413.40	248.00	766.70
		±8.91	±37.83	±31.60	±1.98	±37.76	±5.59	3±29.41	±68.73	±24.47	±6.93
	3-4 week	152.15	294.2	633.85	301.60	633.10	171.85	216.5	502.0	296.40	735.50
		±23.40	±16.26	±19.30	±28.43	±45.25	±8.70	±42.71	±44.12	±37.34	±22.20
2015	1-2 week	191.05	194.95	465.30	250.15	761.65	211.75	133.25	420.35	223.30	782.25
		±46.88	±39.39	±35.21	±45.75	±52.52	±26.52	±3.89	±1.91	±6.22	±8.41
	3-4 week	107.90	283.55	591.40	366.55	599.25	178.25	196.95	509.60	281.10	718.95
		±9.24	±27.51	±58.83	±28.21	±45.12	±32.60	±46.88	±48.65	±47.23	±6.30

Conclusions

The yield of dry matter for the period between the stage without generative stems until the emergence of generative stems increases 2-3 times and is higher in the temporary pasture compared to that of the permanent pasture. The proportion of legume components and content of CP is higher in the temporary pasture in the first year and decreases over the next two years, the proportion of legumes is aligned with that of the permanent pasture. The enzymatic digestibility is higher in the stage without generative stems and decreases by 5 to 21% in stages with generative stems in the temporary pasture and by 5 to 8% in the permanent pasture.

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Impact of long-term fertilization on crude protein fractions of lucerne forage in the first cut

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Abstract

Effects of fertilization on lucerne (*Medicago sativa* L.) forage yield and quality have been widely investigated; however, there are no published studies on the effect of long-term indirect fertilization on crude protein (CP) fractions. The main objective of the present research was therefore to investigate differences in CP fractions under different combinations of mineral (control vs NPK) and organic (control vs manure) fertilization in long-term experiment conducted from 1955 in Ruzyně. Organic and mineral nitrogen fertilization was applied through crop rotation, but not directly at lucerne stand. In 2014, forage was sampled at the bud stage in the first cut in the second harvest year. Manure application without mineral fertilization reached the highest CP content in the forage (248 g kg^{-1} DM). Non-protein fraction A was reduced to 301 g kg^{-1} CP, whereas true protein fraction B_2 was increased to 531 g kg^{-1} CP in the treatment with NPK and manure fertilization. Organic fertilization showed tendency to lower fraction B_1 and indigestible fraction C. This research shows that fertilization can affect lucerne N utilization by ruminants through significant differences in CP fractions.

Keywords: *Medicago sativa*, phosphorus, potassium, manure

Introduction

Protein degradability in forage legumes is of global importance because utilization efficiency of forage has economic and environmental consequences. Rumen protein degradation and the resulting imbalance between carbohydrate and protein supply leads to lower N-use efficiency by ruminants (Broderick, 1995). Increasing the amount of protein that escapes from the rumen could benefit ruminant nutrition and improve the economics of the dairy industry (Chen *et al.*, 2009). Among investigated factors affecting protein fractions include plant species, plant part, harvest maturity, weather condition, and/or stand structure (Hakl *et al.*, 2015). Fertilization belongs to important factors affecting plant growth. The impact of lucerne fertilization has traditionally been focused on forage yield (Vasileva and Kostov, 2015) or forage nutritive value (Lissbrant *et al.,* 2009). Only little attention has been paid to effect of fertilization on the potential of protein degradation. Therefore, this paper presents differences in CP fractions under different combinations of mineral control vs NPK and organic control vs manure fertilization in long-term experiment.

Materials and methods

The Ruzyně Fertilizer Experiment (RFE) was established on a permanent arable field in 1955. The RFE is a large scale experiment consisting of five field strips. Each field strip consists of 24 fertilizer treatments replicated four times and arranged in a complete randomized block design (96 individual plots). The individual plot size is 12×12 m. Analysis of lucerne crude protein fractions response to fertilization management was carried only in strip number IV (first cut, second harvest year) with a 9-year crop rotation system. Four contrast treatments were evaluated in this experiment: Treatment 1 – Control without fertilizers (NIL), Treatment 2 – Mineral fertilizers (MIN), Treatment 3 – Organic fertilizer (ORG), Treatment 4 – Mineral and organic fertilizers (MIN+ORG) (Table 1). Farmyard manure was applied each autumn before planting of sugar beet and potatoes in rates 21 and 15 t ha^{-1}, respectively.

Mineral N was not applied during the cultivation of lucerne. P and K fertilizers were applied in each autumn. Treatment description and average annual rates of N, P and K is given in Table 1.

In 2014, at the late bud stage in the first cut, lucerne biomass was clipped from an area of 0.125×0.5 m in the three replicates. The plant density (PD, m^2), number of all stems (stem density, SD, m^2), maximal stem length (MSL, cm) and leaf weight ratio (LWR, g kg^{-1}) were determined in the clipped fresh samples. Within three hours of clipping, separated samples were oven dried at 60 °C to constant weight. All dried forage samples were milled to pass through a 1 mm screen and analysed for CP fractions. These analyses were carried out by an accredited laboratory (EKO-LAB Žamberk) according to Licitra *et al.* (1996) and protein content was fractionated into A and B_1 (soluble fractions), B_2, and insoluble fractions B_3 and C. Results of stand traits and CP fractions were analysed by one-way and two-way ANOVA with interactions using Statistica 12.0.

Results and discussion

Fertilization treatments significantly influenced lucerne stand traits where increasing rates of applied nutrients continuously increased MSL whilst LWR was reduced (Table 2), independently on form of applied nutrients. Leaves are generally recognized for their excellent forage nutritive value contrast to lucerne stem, therefore reduction of LWR is in accordance with contrasting responses of yield and forage nutritive value described by Lissbrant *et al.* (2009).

Table 1. Average annual rates of N, P and K for fertilization treatment and lucerne stand structure traits in the first cut.[1]

Treatment	Fertilization		Stand structure			
	Organic N:P:K kg ha^{-1}	Mineral N:P:K kg ha^{-1}	Plant density pcs m^{-2}	Stem density pcs m^{-2}	Maximal stem length cm	Leaf weight ratio g kg^{-1}
NIL	0:0:0	0:0:0	168	681	58^{a}	426^{a}
MIN	0:0:0	91:31:146	130	849	68^{ab}	397^{ab}
ORG	30:8:20	0:0:0	177	700	68^{ab}	404^{ab}
MIN+ORG	30:8:20	91:31:146	149	788	77^{b}	381^{b}
P-value			0.317	0.263	0.001	0.040

[1] Different letters indicate statistical differences between treatment for Fisher LSD, α=0.05.

Table 2. Effect of fertilization treatments on concentration of crude protein (CP) and CP fractions (A, B_1, B_2, B_3, C) in lucerne forage.[1]

Treatment	CP (g kg^{-1} DM)	CP fraction (g kg^{-1} CP)				
		A	B_1	B_2	B_3	C
NIL	221^{a}	328^{ab}	68^{a}	509^{ab}	15	80
MIN	222^{a}	418^{a}	75^{a}	403^{a}	24	80
ORG	248^{b}	401^{ab}	19^{b}	492^{ab}	21	67
MIN+ORG	218^{a}	307^{b}	65^{a}	531^{b}	23	73
P – organic	0.178	0.585	0.032	0.128	0.660	0.063
P – mineral	0.100	0.942	0.046	0.342	0.318	0.536
P – organic×mineral	0.083	0.024	0.119	0.061	0.540	0.493

[1] Different letters indicate statistical differences between treatment interaction for Fisher LSD, α=0.05.

ORG treatment reached the highest CP concentration, in spite of only average LWR value in this treatment. It seems that LWR alone is not exclusive predictor for forage CP concentration because a change of CP concentration in separate leaves and stems could also play an important role. Regarding to CP fractions, A, B_1 and B_2 were significantly influenced by fertilization treatments. Fraction C showed a tendency to be lower at manure application, whereas B_3 was stable across treatments. Manure application substantially reduced concentration of soluble CP fractions which could have a positive effect on CP utilization efficiency.

According to Hakl *et al.* (2015), plant morphology was important factor affecting about 75% of CP fractions variability in lucerne. As a contrast to stems, leaves had the highest proportion of true protein (fraction B) and significantly lower soluble and insoluble N fractions. Chen *et al.* (2009) investigated individual proteins from lucerne and found that the largest proportion were photosystem proteins, such as chains or subunits of ribulose bisphosphate carboxylase, which were relatively resistant to digestion after 120 minutes. This matches the higher proportion of fraction B_2 found in the leaves. In spite of the relationship between plant morphology and CP fractions, present results show that differences between fertilization treatments did not fully correspond with changes in stand structure traits. The most suitable CP profile was observed in MIN+ORG treatment with the highest MSL and the lowest LWR values. Contrast to it, the least suitable CP profile with the highest concentration of A and C fraction was recorded in MIN treatment under similar stand structure traits.

Conclusions

This research has shown that effect of long-term indirect fertilization influenced significantly concentrations of lucerne CP fractions. It is possible to conclude that combination of organic and mineral fertilization provided the most suitable CP profile whilst alone mineral NPK fertilization produced forage with the highest ratio of N soluble fraction. Described differences among fertilization treatments could not be simply explained by observed changes in stand structure in particular treatment. It supports the idea about changes of CP fractions within leaves and stems under different fertilization management.

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Effects of harvesting red clover/ryegrass at different stage of maturity on forage yield and quality

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Abstract

Total nitrogen concentrations and digestibility of perennial ryegrass (*Lolium perenne*; PRG) and red clover (*Trifolium pratense*; RC) decline with increasing maturity. However, whilst harvest date is negatively correlated with silage digestibility, it is positively correlated with herbage yield. Currently, there is a lack of information on the ideal cutting stage for mixtures of red clover grown in combination with ryegrass. An experiment was conducted to examine the effects of increasing harvest date on growth stage, dry matter yield, species ratio, forage composition and digestibility of red clover (cv. AberClaret) and perennial ryegrass (cv. AberMagic) grown as a mixed sward. First cut silage was harvested at weekly intervals on 5 occasions from early May to early June, and second cut silage was harvested at weekly intervals on 5 occasions from mid-June to mid-July. Forage nitrogen, neutral detergent fibre and digestible organic matter in the dry matter concentrations were affected by harvest date in both cuts and were significantly correlated with the growth stages of both PRG and RC. Overall, the growth stage of RC was a better indicator of sward forage quality than growth stage of PRG especially in the second silage cut.

Keywords: *Lolium perenne*, *Trifolium pratense*, growth stage, nitrogen, digestibility

Introduction

Crude protein concentrations and digestibility in grass and red clover decline with increasing maturity. Therefore, harvesting early will produce forage for ensiling with the highest CP concentrations and digestibility. For red clover, the different stages of maturity can be determined using the formula and phenological-staging scheme described by Ohlsson and Wedin (1989). Ideally, legumes for ensiling should be harvested at the stage of maturity that maximises nutritional parameters while ensuring adequate sugars for fermentation.

Typically, for grass silage, the decision on when to harvest is usually taken according to grass heading date. For red clover, protein concentrations decline with increasing maturity during first and second cuts for silage production, with King et al (2012) finding CP levels were highest in red clover harvested at early-bud stage during primary growth. However, currently, there is a lack of information to assist farmers growing grass in combination with red clover on whether to cut depending on the stage of growth of the grass or the stage of growth of the clover. Whilst date of harvest is negatively correlated with silage digestibility, it is positively correlated with herbage yield. There are numerous studies which show that increasing silage digestibility increases silage DM intake. The aim of this experiment was to determine the effects of stage of maturity at harvest on the protein concentration, protein yield, digestibility and DM yield when ryegrass/red clover swards are cut for ensiling.

Materials and methods

Three plots (13.5×11 m) of red clover/perennial ryegrass (cv. AberClaret and AberMagic) were established at sowing rates of 10 and 20 kg ha^{-1} respectively in June 2013. In 2014 half of each plot was evaluated during the 1st cut and the second half during 2nd cut. For the 1st cut, areas of the subplot were harvested at weekly intervals on 5 occasions from early May to early June. The second half of each plot was cut on the 14 May in preparation for 2nd cut evaluation which was harvested at weekly intervals on 5 occasions from mid-June to mid-July. All plots were harvested at 8 cm height using a Haldrup plot harvester. Dry matter (DM) and botanical composition was determined by oven drying at 100 °C. Aliquots of fresh forage were freeze dried prior to determination of total nitrogen (TN), neutral-detergent fibre (NDF) and *in vitro* digestible organic matter in the DM (DOMD) contents. In addition, at each harvest date 50 RC and 50 PRG stems were cut at ground level on areas adjoining each harvested area and growth stage (GS) of each stem was scored as described by Ohlsson and Wedin (1989) for RC (0-9) and Moore *et al.* (1991) for PRG (0-4.9). Mean growth stage by weight was calculated for each forage in each plot as the sum of forage growth score multiplied by the stem dry weight for each score category divided by the total dry weight of stems sampled. Yield, composition and GS data were analysed by one-way ANOVA according to the randomised block design and with effects of harvest date partitioned into linear and quadratic contrasts. Spearman's rank correlation coefficients were used to compare (Diedenhofen and Musch, 2015) the strength of association between sward composition and growth stages of PRG and RC.

Results and discussion

DM yield increased linearly ($P<0.001$) with increasing harvest date in both cuts (Table 1 and 2). This yield increase could be attributed to both forages in 1st cut but only to RC in 2nd cut. An increased contribution of RC to DM yield was observed between 1st and 2nd cut, with RC contributing c.60 and c.90% of DM yield respectively but the RC:RG ratio was not affected significantly by harvest date within cut. In 1st cut, TN concentration decreased curvilinearly ($P=0.021$), NDF concentration increased curvilinearly ($P=0.043$) and DOMD decreased linearly ($P<0.001$) with increasing harvest date. In 1st cut, the growth stage of both PRG and RC increased ($P<0.05$) with increasing harvest date reaching maxima of 3.3 and 3.6 respectively. In 2nd cut, TN concentration decreased curvilinearly ($P=0.023$), NDF concentration increased linearly ($P<0.001$) and DOMD decreased linearly ($P<0.001$) with increasing harvest date. In 2nd cut, PRG GS only reached 2.2 while the RC GS reached 7.2. The GS of RC increased with increasing harvest date during both cuts but GS changes were less apparent for PRG particularly during 2nd cut when RC dominated the standing sward. Forage composition was significantly correlated ($P<0.01$) with forage growth scores (Table 3). Correlation coefficients with composition were significantly higher ($P<0.05$) for RC than PRG in 2nd cut but the smaller difference observed between forage species for 1st cut was not significant. The relationship between DOMD and RC GS was linear but for TN and NDF the relationship was nonlinear rather than linear as reported by Ohlsson and Wedin (1989).

Table 1. Yield, growth stage and composition of 1st cut forage.

	Harvest date					s.e.m	Prob	Linear	Quadratic
	7 May	14 May	21 May	28 May	4 June				
Yield (kg ha^{-1})									
Total DM	3,299 ^{a}	4,557 ^{b}	4,805 ^{b}	5,684 ^{b}	6,000 ^{b}	321.2	0.003	<0.001	0.327
PRG DM	1,044	1,544	1,996	2,193	2,309	284.2	0.068	0.008	0.363
RC DM	2,161 ^{a}	2,970 ^{b}	2,797 ^{b}	3,486 ^{c}	3,685 ^{c}	141.4	<0.001	<0.001	0.519
RC %	65	65	58	62	61	3.4	0.420	0.175	0.325
Growth stage									
PRG	2.1 ^{a}	2.6 ^{b}	2.8 ^{c}	3.1 ^{d}	3.3 ^{d}	0.06	<0.001	–	–
RC	2.1 ^{a}	2.4 ^{b}	2.7 ^{c}	3.3 ^{d}	3.6 ^{e}	0.07	<0.001	–	–
Composition (g kg^{-1} DM)									
Nitrogen	29.8 ^{b}	24.1 ^{a}	22.2 ^{a}	21.2 ^{a}	20.3 ^{a}	0.98	<0.001	<0.001	0.021
NDF	363 ^{a}	430 ^{b}	462 ^{bc}	480 ^{bc}	495 ^{c}	13.3	<0.001	<0.001	0.043
DOMD	714 ^{c}	687 ^{bc}	666 ^{b}	625 ^{a}	616 ^{a}	10.0	<0.001	<0.001	0.773

^{a,b,c,d} means differ ($P<0.05$) based on the Student-Newman-Keul's method for multiple comparisons.

Table 2. Yield, growth stage and composition of 2nd cut forage.

	Harvest date					s.e.m.	Prob	Linear	Quadratic
	17 June	24 June	30 June	8 July	15 July				
Yield (kg ha^{-1})									
Total DM	3,248 ^{a}	4,925 ^{b}	5,383 ^{b}	6,461 ^{c}	7,265 ^{c}	270.0	<0.001	<0.001	0.202
PRG DM	453	823	488	495	452	172.2	0.534	0.570	0.478
RC DM	2,788 ^{a}	4,089 ^{b}	4,890 ^{c}	5,963 ^{d}	6,801 ^{e}	207.2	<0.001	<0.001	0.260
RC %	86	83	91	92	94	2.9	0.128	0.027	0.920
Growth stage									
PRG	1.7 ^{a}	1.9 ^{ab}	2.0 ^{ab}	2.4 ^{b}	2.2 ^{ab}	0.12	0.021	–	–
RC	2.6 ^{a}	3.6 ^{b}	4.2 ^{c}	5.8 ^{d}	7.2^{e}	0.16	<0.001	–	–
Composition (g kg^{-1} DM)									
Nitrogen	33.2 ^{c}	26.4 ^{b}	25.1 ^{ab}	22.5 ^{ab}	21.5 ^{a}	1.05	<0.001	<0.001	0.023
NDF	333 ^{a}	380 ^{b}	403 ^{b}	467 ^{c}	513 ^{d}	7.7	<0.001	<0.001	0.400
DOMD	720 ^{e}	672 ^{d}	651 ^{c}	605 ^{b}	546 ^{a}	5.7	<0.001	<0.001	0.120

^{a,b,c,d} means differ ($P<0.05$) based on the Student-Newman-Keul's method for multiple comparisons.

Table 3. Spearman rank correlation (n=15) between growth stage and forage composition.

			Composition (g kg^{-1} DM)		
		RC GS	Nitrogen	NDF	DOMD
1st Cut	PRG GS	0.943***	-0.843***	0.854***	-0.893***
	RC GS	–	-0.904***	0.879***	-0.961***
2nd Cut	PRG GS	0.736**	-0.575**	0.689**	-0.768***
	RC GS	–	-0.850***	0.982***	-0.968***

**, ***; Coefficient differs from 0, $P<0.01$ and $P<0.001$ respectively.

Conclusions

In mixed PRG/RC swards the growth stage of RC is a better indicator of sward forage quality than GS of PRG, especially in the second cut when RC was dominant.

Acknowledgements

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Effects of sainfoin on silage protein when ensiled in combination with either ryegrass or lucerne

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Abstract

Sainfoin (*Onobrychis viciifolia*) is a tanniferous legume, used as a high-protein forage for ruminant livestock. Protein-tannin complexes form at pH range 3.5-7.0, thereby tannins have the potential to reduce protein degradation during ensiling. Polyethylene glycol (PEG) has the ability to bind strongly to tannins and inhibit tannin-protein complex formation. This effect was utilized to study if condensed tannins decrease protein degradation, with PEG shown to affect protein-tannin complex formation in sainfoin silage. The aim of the experiments presented here was to study the effect of using sainfoin when ensiled in combination with other forages. The first experiment determined the effects of sainfoin when ensiled in combination with ryegrass in different proportions. The second experiment determined the benefits of sainfoin tannins on protein degradation when sainfoin was ensiled in combination with lucerne in different ratios, and either with or without PEG. When ensiling ryegrass with sainfoin, silage soluble-N concentrations decreased linearly with increasing proportion of sainfoin. When ensiling lucerne with sainfoin, silage soluble-N concentration decreased linearly with increased sainfoin inclusion in the absence of PEG, but in the presence of PEG it was unaffected by sainfoin proportion.

Keywords: *Onobrychis viciifolia*, *Medicago sativa*, tannin, polyethylene glycol, nitrogen, legumes

Introduction

Sainfoin is a tanniferous legume used as a high-protein forage for ruminant livestock and its moderate-to-high condensed tannin (CT) content has the potential to improve protein utilisation within ruminant systems. Protein-tannin complexes form at pH range of 3.5-7.0, thereby tannins can reduce protein degradation during ensiling (Albrect and Muck, 1991). Previous work has shown that the tannins reduced protein breakdown when sainfoin was ensiled as a pure forage (Lorenz *et al.*, 2010). Other studies on red clover and lucerne bi-crops have shown the benefits of ensiling these two forages together (Marley *et al.*, 2003), due to the benefits of polyphenol oxidase in the red clover protecting the lucerne protein from degradation during ensiling (Jones *et al.*, 1995). Polyethylene glycol (PEG) has the ability to bind strongly to tannins and inhibit tannin-protein complex formation. This effect was utilized to study if condensed tannins decrease protein degradation, with PEG shown to affect protein-tannin complex formation in sainfoin silage (Lorenz *et al.*, 2010). The aim of the research presented here was to study the effect of using sainfoin, a tanniferous legume, when ensiled in combination with other forages. The findings of two separate experiments are presented. The first experiment determined the effects of sainfoin when ensiled in combination with ryegrass in different proportions. The second experiment determined the benefits of sainfoin tannins on protein degradation when sainfoin was ensiled in combination with lucerne in different ratios, and either with or without PEG.

Materials and methods

Sainfoin (SF; Cotswold Common), lucerne (LUC; cv. Timbale) and hybrid ryegrass (HRG; cv. AberEcho) were established in field plots during 2013.

Experiment 1: On 16 June 2014, 1st cut SF at growth stage (GS) 6.7 (i.e. late flowering) and 2nd cut HRG were harvested and wilted for 42 h. Forages were then chopped and treated with a silage inoculant (Ecosyl

100, Volac Ltd, Royston, Herts., UK; 1×10^6 cfu g^{-1}) before ensiling in triplicate 1.5 l glass laboratory silos as mixtures of SF and HRG containing proportionally 0, 0.25, 0.5 and 0.75, 1 SF on a dry matter (DM) basis.

Experiment 2: Second cut SF and 3rd cut LUC (GS 4.8 and 4.4 respectively, i.e. early flowering) were harvested on the 12 August 2014 and wilted for 42 h. Forages were then chopped and treated with an inoculant as above. Mixtures of SF and LUC with 0.1, 0.3, 0.5, 0.7 and 0.9 SF on a DM basis were ensiled in triplicate silos after the addition of water (-) applied at the rate of 300 g kg^{-1} DM of forage or PEG (+; MW 3350; Sigma, Gillingham, Dorset, UK) applied as 90 g PEG + 210 g water kg^{-1} DM of forage. Individual forages at ensiling were analysed for freeze-dried DM (FD DM), and total nitrogen (TN) content. Silages were analysed for FD DM, TN, ammonia-N (Amm-N), buffer soluble N (Sol-N). Data from Experiments 1 and 2 were analysed by one-way and two-way ANOVA's respectively with effects of SF proportion partitioned into linear (Lin) and quadratic (Quad) contrasts and residual deviations (Dev).

Results and discussion

At the point of ensiling TN content was similar for sainfoin and HRG but higher in LUC than in sainfoin (Table 1).

In Experiment 1, SF ensiled at a ratio of 0.25:0.75 with HRG had a higher TN concentration than other HRG/SF combinations despite the forages having similar TN contents at ensiling. (Table 2). Soluble-N concentration (g kg^{-1} TN) decreased linearly ($P<0.001$) with increased SF inclusion reflecting lower N solubility due to the tannins in SF.

In Experiment 2, the addition of dry matter as PEG increased silage DM from 328 to 355 g kg^{-1} ($P<0.001$) (Table 3). As expected from the forage composition, Total-N decreased with increased SF inclusion and was diluted by PEG addition. Ammonia-N (g kg^{-1} TN) decreased linearly with increased SF inclusion

Table 1. Forage composition at ensiling (n=3).[1]

Analyte	Experiment 1				Experiment 2			
	SF		HRG		SF		LUC	
	Mean	s.d.	Mean	s.d.	Mean	s.d.	Mean	s.d.
FD DM (g kg^{-1})	409	3.7	397	6.2	368	5.9	375	0.9
TN (g kg^{-1} DM)	22.2	0.73	22.6	0.35	27.4	0.99	35.7	1.51

[1] FD DM = freeze-dried dry matter; TN = total nitrogen.

Table 2. Experiment 1 SF/HRG silage composition after 125 days ensiling.[1,2]

	Proportion of Sainfoin					s.e.m.	Prob	Prob		
	0.0	0.25	0.5	0.75	1.0			Lin	Quad	Dev
FD DM (g kg^{-1})	383.4	388.0	390.8	390.9	391.2	2.58	0.238	0.047	0.272	0.963
TN (g kg^{-1} DM)	25.2^a	26.7^b	24.7^a	23.6^a	23.7^a	0.45	0.004	0.002	0.310	0.018
Amm-N (g kg^{-1} TN)	66.7	63.4	60.4	75.0	65.5	5.64	0.475	0.617	0.805	0.226
Sol-N (g kg^{-1} TN)	673.5^b	609.9^a	594.3^a	574.8^a	561.8^a	14.01	0.002	<0.001	0.093	0.577

[1] FD DM = freeze-dried dry matter; TN = total nitrogen.

[2] a,b,c within rows differing superscripts indicate means differ ($P<0.05$) based on Student-Newman-Keul's test.

Table 3. Experiment 2 SF/LUC silage composition after 113 days ensiling.[1,2]

	PEG	Proportion of Sainfoin (SF)					Mean	Prob			s.e.m.[3]
		0.1	0.3	0.5	0.7	0.9		SF	PEG	SF×PEG	
Total-N	–	37.2	35.6	33.4	33.3	30.4	34.0	<0.001	<0.001	0.086	0.38
(g kg^{-1} DM)	+	34.9	32.8	31.7	30.1	29.3	31.8	(Lin <0.001)			
	Mean	36.0^{e}	34.2^{d}	32.5^{c}	31.7^{b}	29.9^{a}					
Amm-N	–	131	135	98	86	78	106	<0.001	0.256	0.112	5.8
(g kg^{-1} TN)	+	128	122	110	102	88	110	(Lin <0.001)			
	Mean	129^{c}	129^{c}	104^{b}	94^{ab}	83^{a}					
Sol-N	–	714^{a}	652^{ab}	$^{A}609^{bc}$	$^{A}565^{cd}$	$^{A}530^{d}$	614	<0.001	<0.001	<0.001	15.2
(g kg^{-1} TN)	+	701	697	$^{B}704$	$^{B}699$	$^{B}675$	695			(Lin <0.001)	
	Mean	708	674	656	632	602					

[1] a,b,c,d within rows values with differing lower case superscripts differ significantly (*P*<0.05).

[2] A,B within columns, values with differing upper case superscript differ significantly (*P*<0.05).

[3] s.e.m applies to interaction means.

and since no difference was observed with PEG inclusion this suggest that the reduction was due to improved fermentation rather than tannin influence. Soluble-N concentration (g kg^{-1} TN) decreased linearly with increased SF inclusion in the absence of PEG but was unaffected by the addition of PEG. These findings show the benefits of ensiling forages in different combinations to utilise the benefits of individual pure silages, as shown by Lorenz *et al.*, 2010, for livestock feed production, as was previously found with other studies into the effects of forage mixtures during ensiling (Marley *et al.*, 2003) due to the benefits of secondary plant compounds found within different forages (Jones *et al.*, 1995).

Conclusions

When ensiling ryegrass with sainfoin, silage soluble-N concentrations decreased linearly with increasing proportion of sainfoin. Ensiling lucerne in combination with sainfoin resulted in a decrease in TN with increased SF inclusion due to the higher TN content of the lucerne at ensiling. Soluble-N concentration decreased linearly with increased SF inclusion in the absence of PEG but was unaffected by sainfoin proportion in the presence of PEG. The findings of both experiments support the hypothesis that using sainfoin in mixtures will result in a lower N degradation during ensiling due to the presence of tannins in sainfoin forage.

Acknowledgements

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Productivity and quality of multicomponent grass swards on three soil types

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Abstract

Field trials were carried out with the aim to study continuous green forage production from grass and legume-grass swards in the stage of intensive growth. The 12 mixed multicomponent swards were developed on sod calcareous, sod-podzolic, sod stagnogley soil and were fertilized with P 78, K 90 and N 0, N60 $_{(30+30)}$, N120 $_{(60+60)}$ kg ha^{-1}. The swards were cut four times during the growing season. The legumes in mixtures with grasses of various growth patterns provided continuous green forage production during the whole summer season. The botanical composition and fertilisation affected the average productivity of the sward (8.32-14.67 Mg ha^{-1} of DM). The ratio of legumes and the interaction between legumes and definite cultivars in legume-grass swards determined the crude protein (CP) content in the total DM yield of each sward as well as the CP content for each component in the sward. The average content of the net energy for lactation (NEL) of mixed stands was 5.88-6.63 MJ kg^{-1} of DM.

Keywords: grass mixture, cutting, fertilization, soil types

Introduction

In Latvia's grassland farming, use of grass-legume mixtures containing four to six species is traditional practice, because swards can secure better persistence and more stable productivity (Adamovics *et al.*, 2006). The increasing role of sustainable grassland-based ruminant systems in Europe highlights the use of sown multi-species swards and stresses the necessity of developing comprehensive studies of the influence of different grassland management strategies in different local conditions (Peyraud *et al.*, 2014). Species selection in mixtures for sown grasslands is very important (Hopkins and Holz, 2006). Efficient use of legume-containing swards contribute to reduction of N fertilisation (Lüscher *et al.*, 2014) and improvement of forage quality (Sarunaite *et al.*, 2012). The objective of this research was to determine the potential of multicomponent swards for making secure forage availability for herbage production in a grazing-cutting system under agroclimatic conditions of Latvia.

Materials and methods

Field trials were conducted at three experimental sites in Latvia on different soil type: sod – calcareous soil (pH$_{KCl}$ 6.7, containing available P – 60 mg kg^{-1}, K – 144 mg kg^{-1}, organic matter content – 24-28 g kg^{-1} of soil), sod-podzolic soils (pH 7.1, the phosphorus and potassium level – 253 and 198 mg kg^{-1}, respectively, and organic matter – 31 g kg^{-1}), sod stagnogley soil (pH$_{KCl}$ 6.33, the phosphorus and potassium level – 93 and 111 mg kg^{-1}, respectively, and organic matter – 23 g kg^{-1}).

At each of the three sites the same 12 mixtures were sown in June 2014 without a cover crop, in three replications, with a 10 m^2 harvest plot size. Multicomponent swards were composed of *Phleum pratensis, Dactylis glomerata, Lolium perenne, Lolium boucheanum, Festulolium, Festuca pratensis, Festuca arundinacea, Festuca rubra, Poa pratensis, Trifolium pratense, Trifolium repens,* and *Lotus corniculatus*, and grouped in four types: mixtures composed only from grasses (G); white clover (*Trifolium repens*) and grass (Tr+G) mixtures; white clover, red clover (*Trifolium pratense*) and grass (Tr+Tp+G) mixtures; bird's-foot trefoil (*Lotus corniculatus*) and grass (Lc+G) mixtures. The following fertilisation treatments were used for all mixtures: P78, K90 and N0, N60 $_{(30+30)}$, N120 $_{(60+60)}$ kg ha^{-1}. Swards were cut four times during the vegetation season. The botanical composition (legumes, grass, herbs) was determined after each cutting. The chemical composition of the plants was determined by the following methods: dry matter (DM) – dried; crude protein (CP) – by modified Kjeldahl; crude fibre (CF), neutral detergent fibre (NDF) and acid detergent fibre (ADF) – by van Soest. The metabolisable energy (ME) was calculated on the basis of the chemical composition of DM using digestibility coefficients and full-value coefficients.

The experimental data were statistically analysed using two-way analysis of variance with 'mixture' and 'fertiliser' as factors; the difference among means was detected by LSD at the 0.05 probability level (Excel for Windows 2003).

Results and discussion

The highest average dry matter yields were achieved on sod calcareous and sod-podzolic soils (13.44 and 13.31 Mg ha^{-1}, respectively). Sward productivity on sod stagnogley soil was lower (12.51 Mg ha^{-1}). The swards containing only grass provided lower average DM yields (8.32 Mg ha^{-1}) at all experimental sites. Mixtures containing two species of legumes (Tr+Tp+G) provided higher average DM yields (15.48 Mg ha^{-1}) on all soil types. Mixtures containing white clover performed better on sod stagnogley and sod calcareous soils, but mixtures with bird's-foot trefoil were best on sod-podzolic soil (Table 1).

The N fertilizer dose increase from 0 to 120 kg ha^{-1} contributed to a significant DM yield increase by 4.44 Mg ha^{-1} for grass-only and by 2.44 Mg ha^{-1} for Lc+G mixture swards on all soil types. For Lc+G mixtures, yield increase can be explained by the highest grass proportion in the sward. Application of N fertiliser did not provide a significant DM yield for the Tr+G and Tr+Tp+G swards. For all legume-containing mixtures, N application negatively affected the proportion of legumes in a sward compared to unfertilised plots (Figure 1).

Table 1. Average dry matter yields of grass-only and grass-legume swards, Mg ha^{-1}.

Soil	Mixtures (F_A)[1]	N rate, kg ha^{-1} (F_B)			
		1. (N0)	2. (N60)	3. (N120)	Mean
Sod-stagnogley	G	5.88	7.89	11.31	8.36
	Tr+G	13.76	13.67	15.19	14.21
	Tr+Tp+G	14.44	14.24	14.91	14.53
	Lc +G	12.18	12.17	14.46	12.94
	$LSD_{0.05}$ F_A= 1.37; F_B= 0.69; F_{AB}= 2.38				
Sod-podzolic	G	6.86	10.10	11.76	9.57
	Tr.+G	12.88	12.64	13.42	12.98
	Tr+Tp+G	14.61	13.96	16.92	15.16
	Lc +G	14.55	14.94	17.01	15.50
	$LSD_{0.05}$ F_A= 1.61; F_B= 0.81; F_{AB}= 2.79				
Sod-calcareous	G	5.00	8.13	7.99	7.04
	Tr+G	15.57	15.95	16.45	15.99
	Tr+Tp+G	16.62	16.80	16.81	16.75
	Lc +G	12.70	14.71	14.49	13.97
	$LSD_{0.05}$ F_A= 1.54; F_B= 0.77; F_{AB}= 2.67				
Mean		12.09	12.93	14.23	13.08

[1] G: grasses; Tr+G: white clover, grass; Tr+Tp+G: white clover, red clover, grass; Lc+G: bird's-foot trefoil, grass.

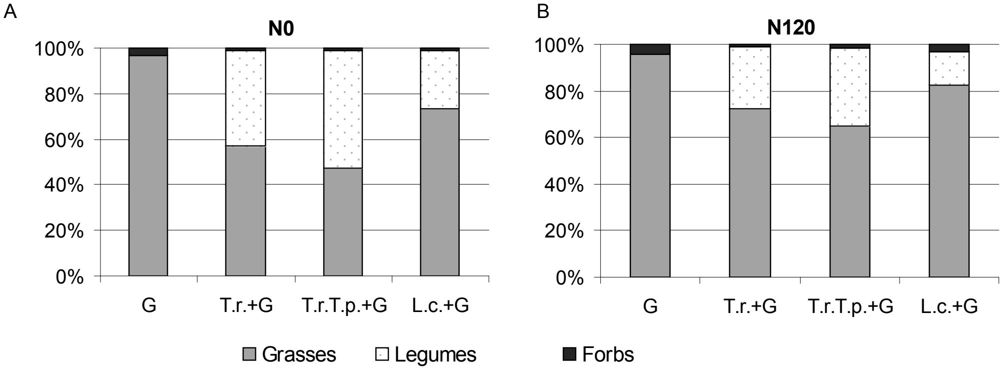

Figure 1. Proportions of the groups of herbage species (%) at two fertilization rates (on average for cuts and three soil types). G: grasses; Tr+G: white clover, grass; Tr+Tp+G: white clover, red clover, grass; Lc+G: bird's-foot trefoil, grass.

The botanical composition of the sward essentially influences the crude protein (CP) and NDF content in dry matter. In grass stands, the average CP content ranged between 133-141 g kg^{-1} DM in grass swards and 162-172 g kg^{-1} DM in legume-grass swards. Our studies showed that mixed grass and legume-grass swards provided CP yields of 1.14-2.69 Mg ha^{-1}.

Average NDF content varied from 401 to 466 g kg^{-1} DM, and ADF content varied from 241 to 282 g kg^{-1} DM. The average NEL content in mixed stands was 5.88-6.63 MJ kg^{-1} DM.

More frequent sward use had promoted the reduction in NDF content in dry matter and had a positive effect on DM digestibility. The coefficient of DM digestibility for grass and mixed legume-grass swards *in vitro* was comparatively high – 66.9-70.1% on average.

Conclusions

Use of legume-containing swards can contribute to reduction in N fertilisation. Mixtures containing white clover performed better on sod stagnogley and sod calcareous soils, but mixtures with bird's-foot trefoil were best on sod-podzolic soil. Mixtures containing white and red clover provided higher average DM yields on all soil types. Application of N fertilizer contributed to decrease in the proportion of legumes in swards.

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Effect of fertilization on yield on permanent grasslands in Serbia

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Abstract

Permanent grasslands are particularly important in hilly-mountainous areas of Serbia, and for many farms they are the only source of forage. These grasslands are generally located on soils with low natural fertility, are of low productivity and have an unsuitable botanical composition. This is because hilly-mountainous grasslands often are overgrown with plant species of low nutritional value. The main means for improving these grasslands consists of adjusting soil fertility, changing the dominance in the vegetation canopy and through good management. This paper presents the results of research obtained during long time period (since 2000) on permanent grasslands, improved by fertilization with different rates and combinations of organic and mineral fertilizers. Results indicate beneficial effect of mineral and organic fertilizers on the dry matter yield on permanent grasslands. The results confirmed that permanent grasslands need to be maintained through mineral fertilizer application, especially application of nitrogen.

Keywords: grassland, fertilizer, yield

Introduction

In hilly-mountainous regions of Serbia, on soil with low natural fertility, natural grasslands are the only source of forage. Grasslands are also very important for men and his environment; they take part in maintenance of biodiversity, carbon fixation from the atmosphere, prevention of erosion, and they can be part of landscape area for recreation and relaxation. For development of livestock production in hilly-mountainous region, grasslands are of paramount importance since they present major or single source of livestock feed (Stošić *et al.,* 2005). Although natural grasslands are floristic complex community, their yields are very low (1.8 Mg ha^{-1} meadows and 0.5 Mg ha^{-1} of pastures) (Stošić *et al.,* 2004). The basic measure for improving of meadows and pastures is fertilization, and knowledge of optimal fertilization is indispensable for better grassland utilization and developing an optimal feeding regime for ruminants (Vučković *et al.,* 2005c).

The use of mineral fertilizers, especially nitrogen, represents a basic measure for improving production of permanent grasslands. Recent research shows that zeolite may reduce the application of mineral N fertilizers on permanent grasslands. Simić *et al.* (2014) reports that the use of cattle manure enriched with natural zeolite can be used as a fertilizer for pastures which contributes to a preservation of nitrogen. In this paper we present the effect of fertilization on yield and herbage quality of permanent grasslands under Serbian agro-ecological and economic conditions.

Materials and methods

This paper presents a review of several articles with results of research on permanent grasslands, improved by fertilization with different rates and combinations of organic and mineral fertilizers.

In an experiment conducted on a natural *Cinosuretum cristati* type meadow in hilly-mountainous region of Serbia (Sjenica-Pester plateau), Vučković *et al.* (2005c) tested different rates of nitrogen fertilizer (0, 40, 80, 120, 160 kg ha^{-1}). An experiment conducted on *Agrostietum vulgare* type meadow (2005-2008) in western region of Serbia included six fertilizer rates (0, 150, 200, 300, 350 and 500 kg ha^{-1} NPK yr^{-1}). An experiment conducted on *Arrhenatheretum elatioris* type meadow (2008-2009) in the hilly mountainous region near Valjevo, also included six NPK fertilizer rates ($N_0P_0K_0$, $N_{50}P_{50}K_{50}$, $N_{100}P_{50}K_{50}$, $N_{100}P_{100}K_{100}$, $N_{150}P_{100}K_{100}$ and $N_{200}P_{150}K_{150}$ kg ha^{-1} yr^{-1}).

Results and discussion

Vučković *et al.* (2005c) reported that nitrogen fertilizer had a positive effect on increasing the yield of a natural *Cinosuretum cristati* type meadow and the maximum DM yield was obtained with 160 kg ha^{-1} nitrogen (on average 4.44 Mg ha^{-1} across two years), which was 2.03 Mg ha^{-1}, or 85.0%, higher than the control. Nitrogen rate, also correlated strongly (r>0.9) and significantly (P=0.01) with chemical composition. There were positive correlation coefficients between N application and crude protein (r=0.999**), ash (0.988**), and fat (0.998**) (P=0.01), and negative correlation coefficient between N application and crude fibre (cellulose, r =-0.998**). These results indicate positive effects of nitrogen application on forage quality.

Association *Agrostietum vulgare* is the highest presented and economically the most important type of permanent grassland in hilly-mountainous region of Serbia. In four years research (2005-2008) on *Agrostietum vulgare* type meadow located in hilly-mountainous region of Serbia, with application of different NPK fertilizer rates (0-500 kg ha^{-1}) Vučković *et al.* (2014) reported that fertilizer $N_{200}P_{150}K_{150}$ (500 kg ha^{-1}) provided the highest yield of dry matter – 8.13 Mg ha^{-1}, but the highest nutrient utilization was obtained with the $N_{100}P_{50}K_{50}$ (200 kg ha^{-1}). The same authors concluded that fertilization has a considerable influence on semi-natural meadows dominated by *Agrostietum vulgare*, regarding their DM yield and quality as well as botanical composition.

Research on *Arrhenatheretum elatioris* meadow type gives similar results as the one on *Agrostietum vulgare*. *Ivaniš et al.* (2013) reported that increase in N, P, K fertilization level resulted in corresponding increases in the quantity of grass dry matter, especially increases in N rate had a favorable effect on DM yield. Results show that maximum average dry matter yield of 7.97 Mg ha^{-1} was achieved with the highest NPK rate (200:150:150 kg ha^{-1}) which present increase of 3.82 Mg ha^{-1} or 1.92-fold more compared with the control.

Conclusions

Levels of mineral and organic fertilization in combination with agro-ecological conditions and type of permanent grassland have strong influences on DM yield. Increasing nitrogen fertilization rate also reduces the weed proportion and increases the appearance of high quality grass species. These research results confirm the necessity of the use of mineral fertilizers, especially nitrogen, on permanent grassland.

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Ground truthing – Evaluation of different methods for estimating yields of grass fields in Norway

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Abstract

Reliable ground truth data are essential for developing good models for estimating grassland biomass or yields using satellite imagery. In the FINEGRASS project we evaluated different methods for measuring grass yields under Norwegian conditions. The methods ranged from approximating average plant height or dropping a disc meter and measuring the stand height, to using a spectrometric leaf area index meter. Measurements were done in diagonal transects of three selected *Phleum*/*Agrostis*-dominated grass fields on different dates in the summers of 2014 and 2015 near the city of Tromsø. All measurements were compared with dry matter from corresponding (0.5×0.5m) cut squares. Disc meter readings produced slightly better linear correlation with cut dry matter yield (R^2=0.88) than measurements of average plant height (R^2=0.82). Variability was much higher for LAI above 3 resulting in poorer correlation with yields (R^2=0.62). The disc meter method may therefore be a suitable alternative for rapid estimates of cultivated grass yields under Norwegian conditions.

Keywords: yield, biomass, disc meter, plant height, leaf area index

Introduction

Good quality of ground truth data is vital when modelling grassland biomass based on remote sensing of spectral reflectance from field canopies. For remote sensing by satellite imagery, a major challenge is covering representative ground sites for large areas (McCoy 2005). Thus, measurements of ground truth data need to be rapid and cost efficient, including both working hours and travelling time between measurement sites. In the project FINEGRASS (http://finegrass.eu) we evaluated different methods for rapid and reliable estimates of cut yields and biomass as ground truth for satellite images (Landsat). This included measuring average plant height with a meter stick, measuring plant stand height using a disc meter, and measuring leaf area index by spectrometry. The suitability of the different methods was evaluated until the first cut at differently managed fields, in order to cover a large range in productivity and developmental stages.

Materials and methods

Measurements and samples for cut yields and total biomass were collected along transects of three different grass fields (of minimum 100×100 m) close to Tromsø in 2014 and 2015. Non-destructive measurements were carried out in the following order: (1) visual estimation of average plant height using a meter stick; (2) average of three leaf area index measurements (LI-COR 2200C); and (3) measuring plant stand height by with a disc meter (30 cm, 200 g). The measurements and samples were collected at biweekly intervals, from growth start to the first cut in the first or second week of July. The grass fields were of mixed composition, sowing ages and cultivation, so as to generate a large range in yields and biomass. The mixtures were dominated by either timothy (*Phleum pratense* L.) (at approximately 50-60% of total coverage) in medium and high productive fields, or common bentgrass (*Agrostis capillaris* L.) (at 45%) in a low productive field. All measurements were carried out within a 0.5×0.5m metal frame (5 cm height), before cutting within the frame for yield (>5 cm stubble height). Dry matter yield of cut samples were weighed after drying in a forced convection oven at 60 °C for 48 hours. Correlations between measurements and dry matter yield were analysed by linear regression using Minitab®16.1.0.

Results and discussion

Using rapid non-destructive measuring techniques on the ground can be a good strategy for optimizing sample size of ground truth data for satellite-based biomass modelling (López Díaz and González-Rodríguez 2003). Especially under Norwegian conditions, with often small interspersed fields and challenging topography/distances between sites. Comparisons of different methods for estimating grass yields up to 600 g m^{-2} indicated that plant stand height measured by disc meter correlated better with yields (R^2=0.89) than did measurements of leaf area index (R^2=0.62) and estimations of average plant height (R^2=0.82). Variability of the disc meter data increased slightly for values above 10 cm (Figure 1). Thus, it is often recommended to replace disc meter measurements above certain stand heights. However, replacement with plant height did not seem to improve accuracy in our case. LAI measurements correlated well with yields up to LAI values of 3 (Figure 2). Above this value, the variability was much higher. This suggests that the sampling protocol needs to include averages of more than three measurements per point

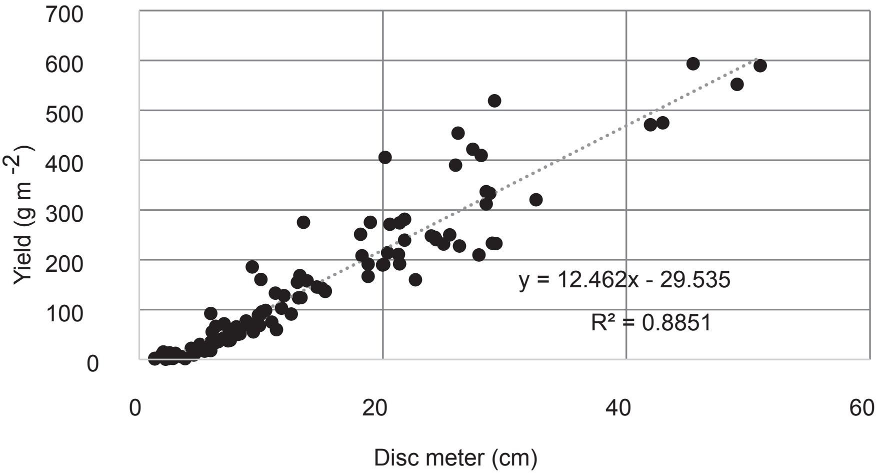

Figure 1. Dry matter yields versus disc meter measurements of timothy and common bent dominated grass fields in Tromsø 2014 and 2015. Total sample size: n=96.

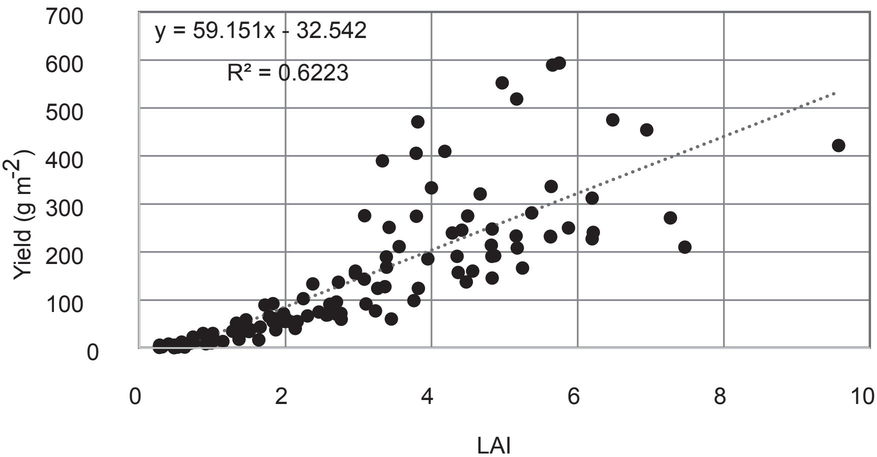

Figure 2. Dry matter yields versus leaf area index (LAI) measurements of timothy and common bent dominated grass fields in Tromsø 2014 and 2015. Total sample size: n=96.

in cultivated grassland, which will add to the time spent per field site. However, LAI may be preferable to disc meter measurements for low LAI values, as disc meter measurements always need to be calibrated for different grass mixtures/species (Bransby *et al.,* 1976). Measuring plant height is the simplest and most rapid method, but the variability of this method was more evenly distributed across the whole range of yields than the other two methods, resulting in less accuracy at low yield levels (below 200 g m^{-2}).

Conclusions

Using disc meter measurements may be a suitable alternative to destructive harvesting for rapid estimations of grass yields over a wide range under Norwegian conditions.

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Theme 3.
Forage potential in ruminant nutrition

Improving utilisation of forage protein in ruminant production by crop and feed management

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Abstract

Protein supplements are usually more expensive than forages or energy supplements, which has increased the interest of improving utilization of forage protein in ruminant production systems. Crude protein (CP) concentration is a poor indicator of productive value of forage protein. The current feed protein evaluation systems estimate the supply of absorbed amino acids (metabolisable protein; MP) that is comprised of microbial protein synthesised in the rumen and ruminally undegraded feed protein (RUP). Microbial protein is the major component of MP in forages. Its supply is mainly regulated by the supply of fermentable energy to rumen microbes. Despite being a smaller component of MP supply there has been more interest to improve forage protein utilisation by increasing the supply of RUP. However, production responses to increased supply of RUP from forages have been disappointing. This can be due to the key role of energy intake in regulating milk protein production and partly due to inherent methodological problems in determining ruminal protein degradability. Analysis of data from production studies indicated forage intake and digestibility are the main forage factors influencing milk protein yield. To improve overall forage protein utilisation reducing the ratio between CP and energy is the most important factor. This can be achieved by avoiding high levels of N fertilisation, optimal harvesting time of grasses, ensuring high fermentation quality and reducing forage CP concentration by using mixtures of whole-crop and grass/legumes silages.

Keywords: metabolisable protein, energy, microbial protein, digestibility, degradability, preservation method

Introduction

Protein utilisation in ruminant production systems is rather low. In large data sets from milk production studies the average N utilisation efficiency (NUE; milk N per N intake) was 26-28% (Huhtanen and Hristov, 2009) and lower in growing cattle. However, the wide ranges in NUE suggests that there is potential for improvements. Protein supplements are usually more expensive than forages or energy supplements. Increased protein prices, environmental concerns of N emissions and ethical aspects of using high quality proteins to ruminants have increased interest of improving utilization of forage protein for ruminants. Because of the high proportion of forage protein to the total protein intake even small improvements in forage protein value can reduce the requirement of supplementary protein and improve overall NUE. Development of new feed protein evaluation systems in 1980's that are based the supply of absorbed amino acids (metabolisable protein; MP) from microbial protein and feed protein form a more comprehensive basis for evaluation of feed protein value. The MP systems have a better potential to optimize protein feeding to meet the requirements of absorbed amino acids with a minimal dietary crude protein (CP) concentration than previous systems based on total CP or digestible CP.

Three different strategies are available to improve utilisation of forage protein by ruminants: reducing forage CP concentration, improving/optimising microbial protein synthesis (MPS) in the rumen and reducing ruminal degradability of forage protein. Both crop and feed management tools can be used to reach these targets. However, these strategies will be not be applied in practise if the production decreases or feed costs increase. Increasing the supply of absorbed amino acids from forages, especially from ruminally undegraded protein (RUP), has been a subject of intensive research. Considerable differences in the predicted supply of RUP from forages have been reported from studies based mainly on the *in situ* (nylon bag) technique or chemical analysis of N fractions. However, these observations have seldom been validated in milk production or digesta flow studies in cannulated animals. Because of the strong negative relationship between dietary crude protein (CP) concentration and overall NUE in milk production, the MP requirements should be met at lowest possible dietary CP concentration without compromising rumen functions, feed intake and performance. The objectives of this paper are to discuss different strategies of improving utilisation of forage protein and how the determined differences in RUP or MP values of forages are realised in production experiments.

Preservation method

Forages are preserved as silage or hay for indoor feeding period. The concentration of DM ranges from about 200 g kg^{-1} in direct-cut silages to about 850 g kg^{-1} in hay. The differences in DM concentration between the harvesting methods have a strong influence on chemical composition of forages that can influence on the protein value. Water soluble carbohydrates are fermented to lactic acid and volatile fatty acids during silage fermentation with the extent depending on ensiling technologies, especially additives and degree of wilting. Nitrogenous fractions in herbages are subjected to proteolysis by plant and microbial enzymes. The changes in carbohydrate fractions can be expected to influence MPS, whereas the changes in N fractions have a greater influence on the supply of MP from RUP.

It has often been claimed that the efficiency of MPS is lower in animals fed silage diets compared to diets based on fresh or dried forages. Microbial protein synthesis regulated mainly by the supply of fermentable energy in the form of ATP to rumen microbes provided that rumen degradable protein (RDP) is not limiting the synthesis. In feed protein evaluation systems MPS is calculated from digestible organic matter (DOM) without any discounts or discounting DOM for substrates that provide no ATP or less ATP than fermented carbohydrates. Ensiling is the most common preservation method of forages. This is at least partly related to silage fermentation products [lactic acids, volatile fatty acids (VFA)] that provide no ATP or less ATP to rumen microbes than water soluble carbohydrates (Chamberlain, 1987). Increased extent of in silo fermentation has been associated with reduced efficiency of microbial N synthesis (Harrison *et al.*, 2003). However, the efficiency of MPS was greater in cattle fed silage-based diets than in those fed hay-based diets (Jaakkola and Huhtanen, 1993). In addition to restricted fermentation of the silage it is possible that the supply of preformed amino acids and peptides stimulated MPS in this study. Holden *et al.* (1994) reported a numerically greater microbial N flow to the duodenum in dry cows fed silage compared with those fed hay. It might be concluded that poor fermentation quality rather than preservation method per se explains the lower efficiency of MPS reported for silage-based diets. In sheep MPS estimated from urinary excretion of purine derivatives MPS was greater for hay and heavily wilted silage than for silages preserved at lower DM concentration (Verbič *et al.*, 1999).

Ruminal protein degradability determined by the *in situ* method has consistently been lower for hay than silage harvested at the same time from the same sward. In the study of Verbič *et al.* (1999) ruminal CP degradability of direct-cut grass silage was on average 15%-units higher than that of hay. Grabber and Coblentz (2009) reported 14, 21 and 5%-units higher proportion of RUP for legume hays than legume silages when determined by the *in situ* method, from protein fractions according to the Cornell Net Carbohydrate and Protein System (Fox *et al.*, 2003) or by a protease method. Ruminal protein degradability was about 5%-units lower for hay-based diets compared with grass silage-based diets in duodenally cannulated cattle (Jaakkola and Huhtanen, 1993), but, because of the higher microbial protein flow with silage-based diets, there was no difference in non-ammonia N (NAN) flow between the forages.

In feed protein evaluation systems the ratio between MP and energy is higher for hay than silage, especially when discounts are made for silage fermentation acids in calculating MP from microbial protein. For example, in the NorFor feed tables (http://www.norfor.info/feed-table/) MP concentration per unit of net energy is about 20% higher for hay than for silage. This suggests that, with dairy cow diets containing 60% forages of total DM intake, preserving forages as hay rather than silage would spare about 2 kg d^{-1} of soybean meal (replacement with grain). The difference between legume hay and silage in RUP determined by the *in situ* method (Grabber and Coblentz, 2009) suggested an even greater protein sparing effect.

Milk production responses are the ultimate test of feeding values. Experimental evidence from a number of milk production studies conducted in Sweden (Bertilsson, 1984) and or Finland (Huhtanen 1994; Shingfield *et al.*, 2002) does not give any support for a greater protein value for grass hay compared to well-fermented grass silages harvested from the same sward. In a series of feeding experiments comparing lucerne (*Medigaco sativa*) hay and silage (Broderick 1995; Vagnoni and Broderick, 1997) the cows fed on hay diets produced more milk protein than those fed silage without protein supplements or lower level of energy supplementation, but with supplemented diets no difference between hay and silage diets were observed. Overall, the differences in milk production responses were closely related to differences in DM intake suggesting that the differences between the forage types were mediated by energy supply. Greater intake and production responses to increased protein (fish meal) or energy supplementation may indicate that silage diets were more limited by the supply of MP than hay diets. In the lucerne studies no silage additives were used, whereas in grass studies silages were preserved with formic acid based additives.

Wilting has decreased ruminal protein degradability determined by the *in situ* method (Tamminga *et al.*, 1992). However, in the analysis of data (Huhtanen and Nousiainen, 2012) from milk production studies (90 diets in 23 comparisons) silage DM concentration had no influence (P=0.90) on milk protein yield when included in a bivariate model with ME intake. In the bivariate model with dietary CP concentration silage DM concentration had a small but significantly negative effect on NUE. These results suggest that production responses to wilting were mediated mainly by increased ME intake and that the lower ruminal protein degradability of wilted silages is not realized as improved milk protein yield or NUE.

Maturity at harvest

It is well-known that harvesting grass silage at an earlier stage of maturity increases CP concentration and improves digestibility, and consequently increase intake and milk production, but decreases NUE. On average, in silage harvest studies (93 diets) marginal efficiency of incremental CP utilization in milk protein production (153 g milk protein/kg increase in CP intake) was greater than the corresponding responses to supplementation of rapeseed meal or expeller (136 and 133) and soybean meal (98) in the meta-analysis by Huhtanen *et al.* (2011). Increased ME supply is the most likely explanation for increased milk protein yield with earlier harvest of forages. In forage harvest studies ME intake explained the variation in protein yield much better than CP intake, and in the bivariate model (ME intake + CP concentration) the effect of CP was non-significant (P=0.43), numerically even negative.

The growth stage of forages affects MP concentration by two different mechanisms. Postponing the harvest decreases digestibility and therefore MPS can be expected to decrease. On the other hand, ruminal CP degradability determined by the *in situ* method tends to decrease with advancing maturity at harvest. When the calculations of MP concentrations are based on *in situ* degradability data (e.g. NorFor; Volden, 2011) the ratio between MP and net energy (NE) decreased with silage NE concentration (Figure 1). This advocates for the MP supply from RUP increasing more with advancing maturity at harvest than the supply of microbial protein decreases. When the calculation of MP is based on a constant CP degradability the MP/NE ratio is independent of silage energy concentration. If the supply of MP relative to energy were lower for early than late harvested silages production responses to supplementary protein would be

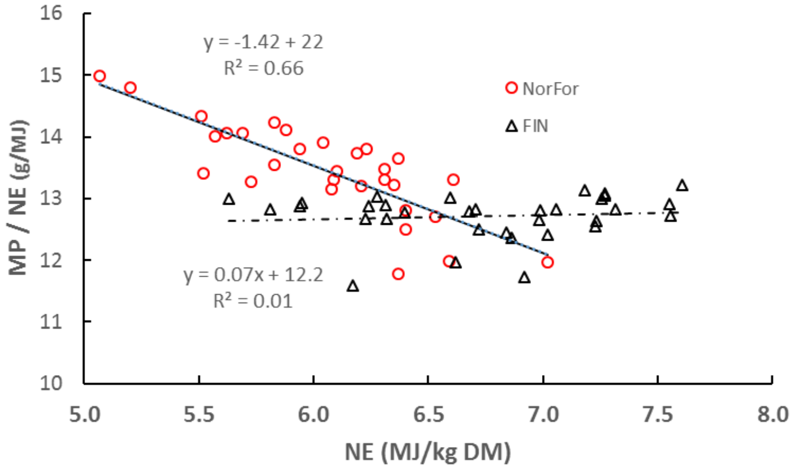

Figure 1. Relationship between silage net energy (NE) concentration and metabolisable protein (MP)/NE ratio according to NorFor feed tables and calculated according to the Finnish system from *in vivo* sheep digestibility data using the constant degradability for all silages (ME was converted to NE using coefficient 0.60).

expected to be greater for diets based on early harvested (high CP) forages compared to late harvested (low CP) forages. However, the analysis of data from protein supplementation studies (Huhtanen, 2013) does not support this, since the marginal milk protein yield responses were not related to CP concentration in the basal diet without supplemental (rapeseed meal or expeller) protein. The results from production studies indicate that the positive effects of improved forage digestibility from earlier harvest and protein supplementation are additive. It can be concluded that earlier harvest of silages increases milk yield mainly due to increased ME intake without any extra benefits from the higher CP concentration.

Nitrogen fertilization

Increasing N fertilization of leys was a widely used strategy in 1970's to increase protein supply for dairy cows. However, this strategy was not challenged by comparing milk production responses to increased CP supply from increased N fertilization and high quality protein supplements. In a later study (Shingfield *et al.*, 2001), increasing fertilization from 50 to 100 kg ha^{-1} increased silage CP concentration from 120 to 150 g kg^{-1} DM but increased CP from silage had no influence on milk or protein yield when no protein supplements were fed despite very low dietary CP and milk urea concentrations (121 g kg^{-1} DM, 1.4 mM). However, when the low CP silage was supplemented with rapeseed expeller to increase dietary CP to the same level as the high CP silage without supplemental protein milk protein yield increased 120 g d^{-1}. The response to rapeseed expeller was the same (115 g d^{-1}) with the high CP silage as with the low CP silage. The results of this study indicate that the cows were responsive to increased protein supply, but the supply of utilizable protein could not be increased by N fertilization. Both silages were well-preserved and had similar DM concentration (340 g kg^{-1}). Yields of milk fat and protein have been decreased in cows given silages prepared from primary perennial ryegrass swards receiving progressive increases in N fertilizer from 72 to 180 kg N ha^{-1} (Keady *et al.*, 1995). Consistent with production studies, increasing N fertilization of grass leys (40, 80 and 120 kg ha^{-1}) had no influence on duodenal NAN flow study in duodenally cannulated animals (Vanhatalo and Toivonen, 1993). It can be concluded that grass fertilization should be optimized according to crop DM yield with no benefits from increased N fertilization in nutritive value.

Extent of in-silo fermentation

Both theoretical calculations and experimental evidence suggest that the efficiency of microbial protein synthesis decreases with increased extent of silage fermentation. This is because lactic acid and VFA provide less or no energy for rumen microbes. Restricting in-silo fermentation by acid applications has increased MPS compared to extensively or poorly fermented silages (Harrison *et al.*, 2003; Jaakkola *et al.*, 2006). Increased concentrations of fermentation acids in silage was negatively associated with milk production in a meta-analysis of data from milk production studies, in which in-silo fermentation was manipulated by additive treatments (Huhtanen *et al.*, 2003). However, when DM intake was included in the model the negative effect of total acid concentration in silage remained significant only for milk fat yield. Discounting fermentable OM for silage acids in calculating the MP value is theoretically correct, but it did not improve the predictions of milk protein yield (Rinne *et al.*, 2009), rather *vice versa*. This may be because silage lactic acid is fermented mainly to propionic acid that the main glucose precursor. Increased supply of glucose can thereby reduce the use of amino acids for glucose production and improve the efficiency of the utilization of MP for milk protein synthesis. As for earlier harvest of silage, greater milk protein yield in cows fed restrictively fermented silages is derived mainly from increased feed and ME intake. Discounting for silage fermentation acids in calculating the MP values can result in greater errors than ignoring this discount. If discounts are made then also the effects of increased glucose supply must be taken into account.

Formaldehyde treatment

The use of silage additives containing formaldehyde has reduced ruminal protein degradability and increased duodenal NAN flow in sheep and cattle (Siddons *et al.*, 1979; Thomson *et al.*, 1981; Siddons *et al.*, 1984). However, protein degradability during 6 or 24 h *in situ* incubations was not influenced by aldehyde treatment (Siddons *et al.*, 1984) indicating that the material disappearing from the bags is not necessarily completely degraded and may escape rumen as NAN. Despite marked increases in duodenal protein flow and uptake of amino acid N from the small intestine, production studies have been less convincing in demonstrating beneficial effects of formaldehyde treated silages. Thomas *et al.* (1981) reported 9 and 5% increases in milk and solid-corrected milk yield with formaldehyde treated silage compare with untreated silage, but they concluded that the responses were mediated via changes in the supply of energy (16% increase in silage DM intake) rather than of protein. In a series of feeding experiments with dairy cows no differences in milk production were observed between cows fed silages treated with formic acid or formaldehyde based additives despite markedly lower soluble N concentrations in formaldehyde treated silages (Ettala *et al.*, 1975). Nagel and Broderick (1992) also observed improved milk and protein yield in dairy cows fed lucerne silage treated with formic acid but not with formaldehyde. Increases in NAN flow with formaldehyde treated silages have been associated with reduced apparent N digestibility. Also OM digestibility has decreased with formaldehyde treatment (Thomson *et al.*, 1981; Siddons *et al.*, 1984). Due to the key role of energy supply in animal performance the benefits of increased protein supply can be masked by reduced energy supply in animals fed formaldehyde treated silages.

Red clover

Red clover (*Trifolium pratense*) has many useful features. As leguminous plant it can fix atmospheric N thereby reducing the requirement of mineral N fertilizers. Mixtures of grasses and red clover have exhibited positive associative intake effects (Huhtanen *et al.*, 2007). In addition, polyphenol oxidase system decreases proteolysis of red clover both during in-silo fermentation and in the rumen. Red clover protein is utilized efficiently in the rumen, and the recovery of incremental protein in the outflow from the rumen was high (Vanhatalo *et al.*, 2009). However, although the NAN flow from the rumen was greater with red clover compared to grass silages (Dewhurst *et al.*, 2003; Vanhatalo *et al.*, 2009), milk protein yield did not increase in these studies indicating a poor utilization of the incremental NAN flow to the small intestine. This can partly be due to lower intestinal digestibility of red clover protein than grass protein. In Finnish digestion trials in sheep fed at maintenance apparent CP digestibility was lower and faecal CP output greater (on average 17 g kg^{-1} DM intake) for red clover silages compared to primary growth grass silages (Figure 2). Although lost from metabolism, increased faecal N excretion with red clover silages is environmentally less harmful than urinary N losses. In the analysis of data from production studies comparing grass and grass/red clover silages (56 diets) forage CP concentration was positively related to milk protein yield in a bivariate mixed effect regression model, but the effect was quantitatively small (0.4 g milk protein per g kg^{-1} increase in forage CP). Vanhatalo *et al.* (2009) speculated that the lack of production response to red clover silage could be related to lower proportion of methionine in omasal N flow compared to grass silage diets. However, the total methionine flow was about 10% greater with red clover diets and the differences in milk protein yield responses were more closely related to intake of digestible OM than to NAN flow or methionine flow.

Prediction model of milk protein yield and N utilization efficiency

Milk production responses is the ultimate test of dietary feeding values. A meta-analysis was conducted to evaluate different forage factors on milk protein yield using a mixed model regression analysis. The dataset comprised 545 treatment means in 180 comparisons of different forage treatments. The MP estimate was estimated according to the Finnish protein evaluation system (LUKE, 2016) that uses a constant protein degradability for each forage. Within a treatment equal amounts of the same concentrate was fed to cows on different forage treatments. A model of forage factors predicting milk protein yield is presented in Table 1. In addition to forage factors, concentrate MP intake was used as a covariate in the model predicting milk protein yield to account for the possible differences in concentrate intake between treatments within a comparison. Forage DM intake and D-value were the most important factors emphasizing the importance of ME intake in regulating milk protein synthesis. Relative to one SD unit variation D-value had almost a 3-fold greater effect on milk protein yield (MPY) than CP

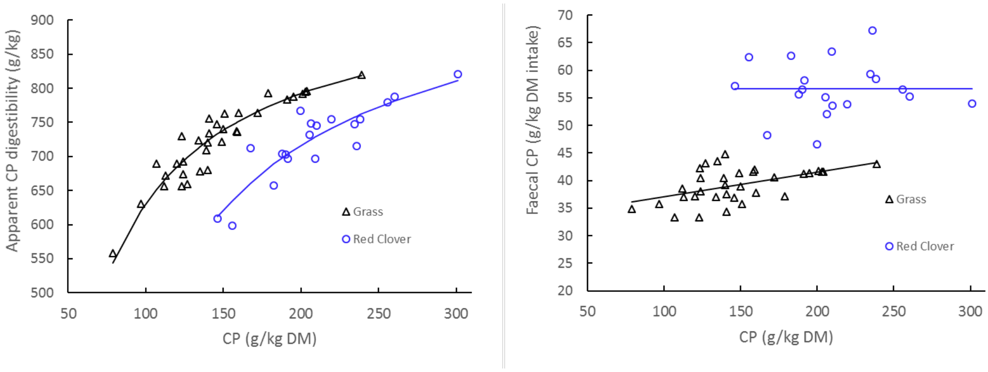

Figure 2. The relationship between CP concentration and apparent CP digestibility (A) or faecal CP output (B) for primary growth grass (n=33) and red clover (n=19) in sheep fed at maintenance level.

Table 1. The best-fit mixed model regression equation of milk protein yield (g day^{-1}) to forage variables (RMSE adjusted for random study effect = 15.0). All parameters were significant at least at *P<0.01.*

Item[1]	Unit	Estimate	SE	SD	Response per SD
Intercept		-277	33.0		
CMP	kg d^{-1}	549	14.8		
DMI	kg d^{-1}	32.5	1.45	1.53	49.7
D-Value	g (kg $DM)^{-1}$	0.42	0.047	40.9	17.4
CP	g (kg $DM)^{-1}$	0.27	0.071	22.4	6.0
Ammonia N	g (kg $N)^{-1}$	-0.14	0.054	21.5	-3.1

[1] CMP = concentrate MP; DMI = forage dry matter intake; D-value = digestible organic matter in DM; CP = crude protein.

concentration. Taking into account the positive relationship between digestibility and intake the relative effect of digestibility compared to CP concentration is even greater. The key role of energy intake on MPY was also demonstrated in the meta-analysis of Huhtanen and Hristov (2009). The effect of forage CP concentration on milk protein yield was small [2.7 g per 10 g (kg DM)$^{-1}$ increase in forage CP], although significant. Of silage fermentation parameters only the effect of ammonia N was significant (*P<0.01*). Silage fermentation acids have a negative impact on MPY, but in this analysis reduced DMI accounts for the observed effects. Milk protein yield tended (*P=0.09*) to increase with the proportion of legume (mainly red clover) silage in forage DM, but the effect was quantitative small (1.7 g d^{-1} per 10% increase in the proportion of legume in forage DM). The effects of forage concentrations of DM and neutral-detergent fibre (NDF), which have been negatively correlated to ruminal *in situ* CP degradability, had no influence on MPY.

Concentration of CP was clearly the most important forage factor influencing NUE within a comparison. Prediction was marginally improved when D-value was included in the model:

$$\text{NUE (g kg}^{-1}\text{)} = 349\pm12.0 - 0.86\pm0.041 \times \text{CP (g kg}^{-1}\text{)} + 0.08\pm0.019 \times \text{D-value (g kg}^{-1}\text{)};$$

RMSE adjusted for random study effect = 5.9.

Additional factors, such as ammonia N, DM concentration or proportion of legume were not significant.

Dilemma between calculated metabolisable protein value and production responses

Ruminal degradability of forage CP has been intensively studied during the last decades by *in situ* incubation technique. These studies have reported wide variations between different forages in ruminal CP degradability, and consequently in calculated supply of RUP and MP. However, there is rather limited evidence that higher RUP concentrations in forages have been converted into increased animal performance. This raises two questions: (1) are there some nutritional limitations in forage RUP, and (2) do current methods determining RUP overestimate the differences between forages.

The meta-analysis of Huhtanen and Hristov (2009) demonstrated the key role of energy supply regulating MPY. Marginal production response to increased supply of feed MP was only 20% of that observed for bacterial MP. They discussed that one reason for the large difference could be overestimation of the range in feed MP and underestimation of that of bacterial MP in the NRC (2001) system. Marginal responses to post-ruminal infusions of casein or amino acids with the same profile as milk (e.g. Doepel *et al.,* 2004) were much smaller (about 0.30) than default values for MP utilization in feed protein evaluation system (0.60-0.65). Because of the inferior amino acid profile of forage proteins compared to casein milk protein and incomplete digestibility of forage RUP a low utilization of incremental MP supply from forage RUP can be expected, especially if not accompanied by increased ME supply. Increases in bacterial MP are accompanied by proportional increases in MP intake, whereas the relationship between feed MP supply and ME intake is much weaker.

Inherent problems of the in situ method can, however, be the main reason for the inconsistency of *in situ* determined RUP and production performance. One assumption of the *in situ* method is that the *a*-fraction immediately disappearing from the bags is completely degraded. However, omasal flow measurements, *in vitro* studies, and studies using ^{15}N-labeled feeds have also shown a considerable flow of feed soluble NAN (SNAN) with peptides being quantitatively the most important component (Choi *et al.*, 2002; Hedqvist and Udén, 2006; Ahvenjärvi *et al.*, 2007; Reynal *et al.*, 2007). Peltekova and Broderick (1996) determined separately the degradability of water soluble and insoluble lucerne protein in *vitro*. Predicted escape was about 20% for both silage and hay soluble protein. Total CP degradability was higher for silage than hay (0.70 vs 0.65), i.e. much smaller than typically observed in studies using the *in situ* method. Consistently, the effect of the proportion of SNAN on MPY was not significant when it was included in a bivariate regression model together with MP intake that was estimated using constant ruminal CP degradability and intestinal digestibility of RUP for forages (Huhtanen *et al.*, 2008). Based on assumptions of the models calculating effective CP degradability from kinetic parameters the regression coefficient for SNAN should have been negative (constant degradability overestimates RUP with increasing SNAN).

Rinne *et al.* (2009) calculated silage AAT values for 397 diets either using constant effective protein degradability (EPD) for all silages or EPD estimated from empirical equations of Yan and Agnew (2004). Their equations were based on *in situ* incubations 136 silages. They used DM, CP, NDF, soluble N and lactic acid/VFA as predictors of silage EPD. Their models explained about 80% of the variation in the *in situ* EPD. Interestingly, ME intake based on constant forage EPD predicted milk protein yield responses better than ME intake calculated using EDP values estimated using Yan and Agnew equations. The coefficients of CP and NDF in the EPD-model were almost opposite to the coefficients used to correct EPD values for microbial contamination. Proportion of microbial N in undegraded residues increases with increased NDF (reduced digestibility) and declining forage CP concentration. It appears that differences in forage EPD determined by the *in situ* technique reflect more variation in microbial contamination than true differences.

Summary and conclusions

The demand of high quality plant protein feeds for human foods and for simple-stomached animals will increase in the near future and so may prices of protein feeds. Because of low marginal efficiency of utilization of supplementary protein in ruminants and increasing prices extensive use of high quality protein supplements for ruminants may be difficult to justify in the future. This has increased interest in improving utilization of forage protein in ruminant production systems. The main focus in improving the utilization of forage protein has been to increase the supply of RUP by decreasing ruminal CP degradability and thereby reducing ammonia N losses from the rumen. Development of ruminal *in situ* techniques in 1970's provided an easy and handy tool to determine ruminal CP degradability. However, there is very little evidence from production studies that reduced in situ degradability has resulted in improved production or NUE. In some cases (e.g. hay vs silage, forage maturity) this can be due to inherent problems of the *in situ* method (microbial contamination, escape of SNAN fraction, initial and secondary particle loss, incorrect passage models). Even when duodenal NAN flow was increased (formaldehyde containing additives, replacement of grass with red clover) production responses have been minimal and usually related to changes in energy intake. In these cases the lack of expected production responses can be related to lower intestinal protein digestibility, non-ideal amino acid composition of forage protein and energy supply becoming the limiting factor. It might be concluded that intensive research on forage protein degradability has not markedly improved the accuracy of evaluation of forage MP value.

The analysis of data from production experiments clearly indicated that energy supply (intake, digestibility) from forages is a driving force of production responses with only marginal effects of CP concentration or quality (silage ammonia content). Predictions of MPY were at least as good when forage MP value was calculated as direct function of D-value compared with a model taking into account CP concentration with constant degradability and digestibility. High digestibility and good fermentation quality are the most important forage factors influencing DM and energy intake, and animal performance, with CP concentration and protein characteristics having only a minor role.

Avoiding high CP concentration is the best strategy for improving efficiency of forage protein utilization. However, this objective may be in conflict with optimizing the economy of milk production. Crop management has a much greater potential to improve forage protein utilization than feed management. Avoiding unnecessary high levels of N fertilization and too early harvest of grass and legume leys is a win-win situation: both production costs and N emissions would decrease. Feeding forage mixtures allows for reducing forage CP concentration without compromising intake and production. Whole-crop silages, especially maize but also barley or wheat silages, have lower CP concentrations relative to digestible OM than legume or grass silages. Although the digestibility of barley and wheat silages is lower than that of grass silages, positive associative intake effects compensates for the lower digestibility and energy intake could be maintained at a lower dietary CP concentration. Including a whole-crop in the crop rotation can also improve the utilization on manure N.

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Grazed grass in the dairy cow diet – how this can be achieved better!

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Abstract

Grassland based systems offer farmers the three pillars of sustainability: economic, environmental and social. Two reasons are proposed for Europe to remerge with a better focus on grazed grass and grazing system efficiency; (1) the economic and demographic situation is rapidly changing in the world, and (2) environment sustainability can no longer be comprised. Economic success in grazing systems is defined by pasture utilisation which, in turn is positively affected by pasture availability and stocking rate (cows/ha) and negatively affected by the amount of feed purchased. Success in this regard will need to build on that principle by growing and utilising more grass per hectare, converting the grass as efficiently as possible into saleable product with a labour and capital efficient system of milk production. The objective of this paper is to outline the reasons for why grazed grass in the cow's diet should be increased and to discuss how such an increase can be achieved.

Keywords: grass, grassland management, sustainability, resilient systems, low-cost feed

Introduction

Negotiations on a major reform of the Common Agriculture Policy (CAP) covering the period 2014-2020 were completed in 2013. The current Multiannual Financial Framework (MFF) for the EU budget fixes budget ceilings for EU agricultural spending up to 2020. In terms of EU policy, stability can be expected as far as 2020. World population is expected to grow by 33% or 2.3 billion people by 2050. Nearly all of this growth is forecast to take place in developing countries. In addition to the projected global population growth, an estimated 3 billon consumers will join the middle classes over the next 20 years, with the middle classes in India and China estimated to reach 50 and 20% respectively of their total populations by 2050. Feeding a world population of 9.1 billion people in 2050 will require increasing overall food production by 70% by 2050 from 2005 levels (DAFM, 2015). Demand for dairy products will continue to expand at a rapid rate through the next decade. Per capita consumption of dairy products in developing countries is expected to increase by 1.2 to 1.9% in the next decade, with the expansion in demand reflecting robust income growth and further globalisation. In contrast, per capita consumption in the developed world is projected to increase by between 0.2 and 0.9%.

Of the grazing livestock population (in LU) in the EU27, 82% are cattle and 14% are small ruminants (sheep and goats), horses making up the remainder. Dairy cows account for 31% and other cows (mainly suckler cows) for 16% of the total LU grazing livestock. Specialist grazing livestock holdings declined faster than specialist field crop holdings (declines of 46 and 33% respectively; Huyghe *et al.*, 2014). All of these findings shows that grazing in Europe is declining even though, when managed well, grass-based dairying can be a very profitable and worthwhile livelihood. Dillon *et al.* (2005) and Shalloo (2009) have shown that key drivers of profitability at farm level in temperate regions are associated with increased grass utilisation. There are a variety of reasons for this decline in grazing including increasing stock numbers per farm, declining labour availability, summer feed deficits, all year round calving, high milk output cows, farm fragmentation, large number of holdings per unit, and mixed farming enterprises (Hennessy *et al.*, 2015).

Maximising the utilisation of grazed grass will have benefits in all systems including economic sustainability and system resilience. High input dairying largely encouraged before the last CAP reform was based on obtaining a high milk price. Since milk quotas and price protection have been dismantled in 2015, high milk price is no longer a guarantee. In the last decades, Europe has regressed in permanent grasslands; this decline needs to be halted as Europe will not be able to sustain its protein needs without these grasslands. The question is can Europe adapt and remerge as a grazing continent in future years?

The objective of this paper is to outline the reasons for and the key aspects of increasing the use of grazed grass in the dairy cow's diet. Much of this work can be applied in many parts of Europe; indeed the authors of the paper have completed many common experiments in their respective nations.

Is grazing interesting for European farmers and consumers?

The grass challenge is now being faced by European farmers. In the past twenty years European production systems have moved away from grass based systems into more high-cost and intensive systems. Now given concerns of the sustainability and economic resilience of such systems, a clear movement is taking place back to grassland systems. Some parts of Europe do not have grass as the main feed source in livestock systems, due to poor climate, unsuitable land types and fragmentation. While concentrate prices have for a large part remained low, parts of Europe have built production systems based on high concentrate feed input. Because of this development, grassland and its optimisation has suffered across Europe. Since the introduction of milk quotas in 1984, the number of dairy cows has declined and milk yield per cow has increased. Many countries have now adopted a flat monthly milk production curve – with year-round calving as the norm. Feed sources have moved from grass to conserved feed stocks, mainly silage from either grass or maize. Such feeds needs to be supplemented with high content protein balancers – adding more costs to the system. Table 1 outlines the main system differences between grass and indoor milk production systems. In many ways these systems are widely different, with grassland systems based on the adoption of more flexible orientated approaches with less focus on output per cow and more focus on low costs. Milk price is now at low levels and many farmers in Europe are producing milk at unsustainable production costs.

The competitive advantage of grass based milk producing nation's lies in their ability to utilize grazed grass as the major feed source. Success in this regard will need to build on this competitive advantage by growing and utilising more grass per hectare, converting that grass as efficiently as possible into saleable product with low capital costs and labour efficient milk production systems.

Table 1. The main differences between grazing and indoor systems.

Grazing systems	Indoor systems
Grazing utilisation based	Conservation based (harvesting/zero grazing)
>200 days at grass	0-100 days at grass
Medium output milk production per cow	High output milk production per cow
Low fixed and variable cost	High fixed and variable cost
Minimum to low building investment	High level building and slurry storage investment
Machinery – minimum/high contractor usage	Machinery-high requirement/little contractor usage
Labour flexibility/Mindset open	Labour routine/Mindset closed
Seasonal workload	Similar workload all year
Resilient system – More consistent margins at irrespective of milk prices	High risk system economically – Exposed at low milk price

Product quality and consumer demand

Dairy products derived from pasture-based systems are considered by consumers to be 'more natural' (Verkerk, 2003) as cows are in a more natural environment which allows the expression of normal behaviours. The effect of dietary regimen on milk composition has received much attention in the past and it is widely accepted that feeding regime has significant effects on milk fatty acid composition with particular emphasis on the health-benefiting unsaturated fatty acid components, particularly conjugated linoleic acid. Feeding regimes that have been studied for their effects on milk include consumption of total mixed rations (White *et al.*, 2001), red clover (Lee *et al.*, 2009), red clover and grass silage (Moorby *et al.*, 2009), fresh alfalfa (Castillo *et al.*, 2006), alfalfa silage (Whiting *et al.*, 2004), fresh forage and marine algae (Glover *et al.*, 2012) and various proportions of fresh grass (Couvreur *et al.*, 2006). In all cases, the provision of fresh grass in the cow's diet has improved milk fatty acid composition.

European consumers have concerns about food quality and safety and tend to view grass based milk production systems as being sustainable, safe and capable of delivering high quality products and multiple ecosystem services (Van den Pol-Van Dasselaar *et al.*, 2014). Citizens mention that they are prepared to pay more for milk from grass based production systems, but dairy companies often indicate that it is difficult to get the associated money from the market. In Ireland, Ornua is the main international marketing source of Irish dairy products. They promote Irish dairy products on the international stage. Most European consumers recognise the Kerrygold brand as an Irish food source and can relate it to dairy cows grazing in their natural environments. The 'Origin Green' trademark is now synonymous with Irish produced products. In the Netherlands the percentage of dairy cattle grazing decreased from 90% in 2001 to 70% in 2013 (Van den Pol-Van Dasselaar *et al.,* 2015). But this decline has now become a societal issue. In 2012, in the Netherlands a voluntary agreement 'Treaty Grazing' was signed by all stakeholders with the aim of stabilising the number of farms practising grazing. Related to this agreement, Dutch dairy companies now provide farmers with higher milk price based on a grazing premium. In France, the PDO products are clearly maintaining their market share; this allows farmers to continually improve the efficiency of their production system. French consumers are very satisfied and readily support these systems largely based on grazing in lowland and mountain areas. Dairy farmers may be missing the bigger picture when grazing is implemented. In general milk solids increase and production costs decline, and if a milk price premium can be generated then more revenues can be generated from the system.

Economy sustainability

Milk price volatility is a new challenge for all milk producing nations. The volatile price is one of the main reasons for why grass based dairying is of interest to nations. Economic efficiency is defined as maximising the returns from a fixed set of resources, e.g. land, labour and capital (McInerney, 2000). In dairy production systems, land is an important fixed resource. The farmer has a choice in terms of the production system he/she adapts based on land availability. For grazing, especially the land availability around the milking platform is vital. Fragmented area increases production costs and reduces the global efficiency of the system. Many studies show that grazed grass is the lowest cost feed for milk production in Ireland (e.g. Dillon *et al.*, 2005; Finneran *et al.*, 2012). As grazed grass is a natural resource, it reduces the requirement for around purchased feed and conserved forage. Dillon *et al.* (2005) showed that total costs of production tend to increase as the proportion of grazed grass in the milk production system declines. French *et al.* (2015) showed that 56% of the variation in milk production costs in Ireland can be explained by the quantity of grass utilised by the dairy herd; each extra ton of grass dry matter (DM) utilised was worth €267 per ton to the farm. Ramsbottom *et al.* (2015) in a recent study of Irish farms found that farms with longer grazing seasons harvested a greater amount of pasture (an additional 19 kg DM ha^{-1} per grazing day per hectare) and greater pasture harvested was associated with increased milk component yield per hectare (58.4 kg of fat and 51.4 kg of protein more per tonne of DM pasture harvested ha^{-1})

and net profit per hectare (€268 ha^{-1} more per tonne of DM harvested). Resilience in a farm business consists of the ability to manage or adapt to change and is a key factor for financial sustainability in a volatile commodity market. Financial success in grazing systems is defined by pasture utilisation, which in turn is positively affected by pasture grown and stocking rate (cows ha^{-1}) and negatively affected by the amount of feed purchased.

Environmental sustainability

One of the key challenges facing agriculture today is centred on the requirement to reduce its environmental losses and impacts. Future milk production systems will need to minimise nutrient losses to water (nitrate, phosphorous) and emissions to the atmosphere (greenhouse gas (GHG), ammonia) so that the production system operated are acceptable by society as a whole (i.e. good animal welfare, preservation of the ecosystem, landscape, biodiversity). The production system must be profitable, afford a good work-life balanced with family life, and provide a good working environment for the farmer and any staff that are directly employed in the business (Figure 1).

Grass based systems are more resource efficient as they use home grown feed stuffs and minimise the requirements for purchased feedstuffs and therefore the resources (area, energy, machinery) associated with those feedstuffs. Total consumption of non-renewable energy is reduced in grass based systems compared to indoor systems (Le Gall *et al.*, 2009). Many studies have been undertaken at country level examining the implications of different production systems on GHG emissions (Casey and Holden, 2005; O'Brien at al., 2012; Schils *et al.*, 2005), eutrophication (Basset-Mens *et al.*, 2009; Benoit *et al.*, 1995; Benoit and Simon, 2004; Briggs and Courtney, 1989) and biodiversity (Atkinson *et al.*, 2005; McMahon *et al.*, 2010; Nitsch *et al.*, 2012; Taube *et al.*, 2014). While all studies use different methodologies and are therefore difficult to directly compare, one key conclusion is evident across all studies: reduced resource utilisation or increased resource use efficiency is associated with increased environmental sustainability.

It is well accepted that there is a high N surplus in grazed grassland due to N fertiliser use, N fixation by legumes (when present) and urine and faecal local deposition by grazing livestock. Permanent grassland acts as a store for N (Brogan, 1966; O'Connell *et al.*, 2003), lowering the risk of N loss to water. In long-term productive grassland soils there is usually net N immobilisation (Jarvis and Oenema, 2000). McCarthy *et al.* (2015) showed that increasing stocking rate, while keeping concentrate input and fertiliser N input constant, increased N use efficiency and reduced surplus N in grass based milk production systems due to increased grass utilisation. Grassland has a high capacity to capture N as grass is present year round and grass is actively growing for a large part of the year (7 to 10 months). With very high stocking rates or in overgrazed situations, N losses can be high because faeces and urine are not evenly distributed over the field during grazing. This leads to more N leaching, more denitrification and more nitrous oxide emissions. Ammonia volatilisation on the other hand is less during grazing. Permanent grassland is ploughed infrequently or not at all, thereby minimising N loss from cultivation.

Social sustainability

Geary *et al.* (2014) reported that spring calving grass based systems had higher net profit per farm than less seasonal calving systems due to lower labour demand. The effect of grazing on labour in year round

Figure 1. The three pillars of sustainability: economic, environmental and social.

calving systems is highly variable. In theory, grazing leads to less labour hours, since the cows graze themselves, and require less management. Other economic and labour efficiency advantages of grazing include reduced reseeding costs (reseeding of grass pastures is not necessary on an annual basis), reduced costs for mechanically harvesting of grass and spreading of slurry, less feed storage and slurry storage costs.

How to improve grazing efficiency

The grazing season needs to be broken down into the different grazing sub-seasons, as in grassland management what happens in one season has direct impacts on the next season. In many respects, the autumn period is the time when paddocks are set up for spring grazing.

Autumn grazing management

In the climatic oceanic regions, characterised with mild winter, the grazing season begins in the previous autumn, i.e. autumn grassland management is one of the main factors influencing grass availability in the following spring. The two main objectives of autumn grazing management are (1) to maximise the proportion of grazed grass in the diet of the dairy cow during this period, and (2) to finish the grazing season with the desired farm cover. Sufficient grass for the remainder of the grazing season can be accumulated by increasing rotation length to greater than 30 days from mid-September. Pre-grazing herbage mass should be maintained below 2,500 kg DM ha^{-1}; if this is exceeded other stock (e.g. dry cows) should be used to graze the paddock(s).

Grazing management practices and climatic conditions dictate that pre-grazing HM should be greater in autumn than spring to facilitate the extension of the grazing season into late autumn. Consequently, pre-grazing HM in autumn is frequently greater than 2,000 kg DM ha^{-1} (>4 cm). Hennessy (2005) reported that autumn swards with a pre-grazing HM of 1,500 kg DM ha^{-1} had a greater proportion of leaf (+0.22) compared to swards with a pre-grazing HM of 2,100 kg DM ha^{-1}. This resulted in a +10 and +48 g kg^{-1} difference in CP (249 g kg^{-1} DM) and DM digestibility (807 g kg^{-1}), respectively. Higher pre grazing herbage masses are targeted in September and October to extend the grazing season, however in Ireland, closing pastures begins from early October.

The key guidelines in Ireland to ensure that there is a supply of spring grass are:
1. Begin closing the farm in rotation from October 10^{th}.
2. Target 60% of the farm to be closed (no more grazing) by November 7^{th}.
3. Complete all grazing by end of November.

The final grazing rotation should commence on October 10^{th} – every paddock grazed from this date onwards should be closed. During the final grazing rotation post-grazing residuals of 100 to 150 kg DM ha^{-1} (4.0 cm) should be targeted to encourage over winter tillering. Each day of delay in closing from October 10^{th} will reduce spring grass supply by approximately 15 kg DM ha^{-1} (O'Donovan, 2000).

Spring grazing

Early spring grazing has a large influence on the success of the total grazing season. Turnout should occur early, and the transition from indoor feeding to grazing should take place early in the spring calved cows lactation. This requires excellent supervision in periods of rainfall and challenging soil conditions. The objective is to graze the paddocks after winter and to prepare high quality regrowth for the second rotation. Positive effects on grass offered and animal performance have been observed by O'Donovan *et al.* (2004) in a study where swards grazed early in spring or not grazed till late spring was compared during the spring/summer grazing season (Table 2).

Table 2. Effect of first spring grazing date and later stocking rate on the grass quality, intake and milk performance in spring (O'Donovan *et al.*, 2004).[1]

First grazing date / later stocking rate	Early grazed / high	Early grazed / medium	Not grazed / high	Not grazed / medium
Grass allowance (>5 cm – kg DM cow^{-1} d^{-1})	12.7	15.9	18.2	21.9
Pre grazing height (cm)	12.2	12.4	14.8	14.2
Leaf proportion (at ground level – % DM)	41	41	39	35
UFL value (1 UFL = 1,700 kcal NEL kg^{-1} DM)	0.99	1.00	0.94	0.94
Crude protein (g kg^{-1} DM)	202	184	171	176
Post grazing height (cm)	4.4	5.0	6.4	6.8
Grass intake (kg DM cow^{-1} d^{-1})	13.9	16.2	16.3	17.0
Milk yield (kg cow^{-1} d^{-1})	20.3	23.9	22.8	23.9
Milk solids (g cow^{-1} d^{-1})	1,356	1,550	1,508	1,554

[1] First grazing occurred in March for the early treatment. The 'not grazed' treatment was grazed for the first time in April. The results presented are those observed during the two later rotations in April and May. DM = dry matter; NEL = net energy lactation; UFL = Unite Fourragère Lait.

Early grazed swards had improved grass quality with a higher proportion of leaf, and consequently a higher nutritive value in energy (UFL) and protein content. The pre grazing height was lower and the reduction of grass allowance in the Early grazed / Medium treatment compared to the Not grazed / Medium treatment affected daily grass intake negatively (-0.8 kg DM), but did not affect daily milk yield and milk solids produced. Compared with the Not grazed/High treatment, with a same grass intake, the grass quality improvement resulted in increased daily milk yield (+1.1 kg). As the grass regrowth structure is more favourable, the post grazing height is lower and the grass is better utilized after an early spring grazing sequence. As observed by Kennedy *et al.* (2006), lax grazing on late spring grazed swards had a cumulative negative effect on grass dry matter intake after four rotations.

Mid-season grazing management

Increasing the number of grazing days is a key priority for increasing the grass production and utilisation on farms. The most important aspect around grazing management is that the farmers must control the feed supply to the grazing herd. The most important aspect of mid-season (April to August) grazing management is to control grass supply. Completing farm cover estimation weekly and assembling the data using the Grass Wedge (Hennessy, 2012) is a simple method to handle and interpret this data and control grass supply. Grass is a moving dynamic system changing the daily supply and careful management especially during periods of high grass growth is needed. A grazing experiment compared three pre-grazing herbage masses (low – 1000 kg DM ha^{-1}; medium – 1,500 kg DM ha^{-1} and high – 2,300 kg DM ha^{-1}) for dairy cows (Table 3) (Tunon, 2013). Daily herbage allowance was 17 kg DM cow^{-1} d^{-1} (>4.0 cm). Grazing at low and medium herbage masses had a positive effect on milk solids yield, as well as on grass utilisation. Continuously grazing at low herbage mass during the grazing season doubled the area required for grazing compared to grazing at high herbage mass and increased the area required by 30% compared to medium herbage mass. Short grazing rotations (<16 days) affects grass production negatively as the sward will rarely achieve the 'three leaf stage'. Another negative effect of the low mass swards was they had to be grazed for 90 min longer daily to achieve a similar intake as medium and high mass swards.

Achieving three leaves on perennial ryegrass tillers is desirable to ensure canopy closure which stimulates high levels of growth. Swards grazed at the medium and high leaf herbage mass both had up to 3 leaves per tiller available at grazing time, while the low mass sward had only 1.5 to 2 leaves per tiller. The

Table 3. The effect of pre-grazing yield on the performance of spring calving dairy cows from April to October.

	Low mass	Medium mass	High mass
Pre grazing yield (kg DM ha^{-1})	974	1474	2319
Pre height (cm)	6.6	9.1	12.4
Post grazing height (cm)	4.0	4.2	4.3
Milk solids (kg cow^{-1})	1.63	1.63	1.58
Dry matter intake (kg cow^{-1})	15.3	16.2	16.2
Grazing time (h)	10.8	9.3	9.3

recommendation is to target pre-grazing yields of 1,300-1,600 kg DM ha^{-1} (>4 cm) during the mid-season period (April to late August) and to graze paddocks to 4-4.5 cm. When pre-grazing yield increases above this the paddock or paddocks should be harvested for round bale silage, closed for a main cut of silage or grazed by non-lactating stock.

Another distinct advantage of this type of grazing system was that offering grazing dairy cows high quality medium herbage mass swards reduced CH_4 emissions per cow per day (282 g CH_4 cow^{-1} d^{-1}) and per kg milk solids (MS) produced (203 g CH_4 kg MS^{-1}) compared to cows grazing high herbage mass swards (+21 g CH_4 cow^{-1} d^{-1} and +26 g CH_4 kg MS^{-1}) (Wims *et al.,* 2010).

Wet weather management

Poaching damage is an associated risk when increasing the length of the grazing season, particularly during times of soil saturation which usually occur in early spring and autumn. Tunon (2013) observed in a free draining soil that DM yield was reduced by 0.30 in badly poached paddocks after damage but cumulative DM yield was not different between undamaged and badly poached treatments (12.7 t DM ha^{-1}). In a heavy soil, annual DM yield was reduced by between 14 and 49%, depending on frequency of poaching and timing. A predominantly perennial ryegrass (PRG) sward on a free-draining soil is resilient to heavy treading damage but a PRG sward on wet soil requires careful management to avoid significant losses in DM production after poaching damage.

Every farm can incorporate grazed grass into the diet of dairy cows through different management strategies. When soil or weather conditions are poor, restricted access to grazing can be practiced (Kennedy *et al.*, 2009, 2014; Pérez-Ramírez *et al.*, 2008). This approach involves turning cows out to grass for a short fixed time period each day. Kennedy *et al.* (2009) showed that spring dairy cows were able to achieve 95% of their daily grass DM intake when provided with access to grass for three hours after morning and evening milking, and milk production was similar compared to cows that were fulltime grazing in the same period. In autumn, Kennedy *et al.* (2014) found no negative effects on milk production of restricted access to grass offered to cows in late lactation. In France, Pérez-Ramírez *et al.* (2008) reported that restricting access time to pasture reduced milk yield and composition in spring and early summer. The practise of restricted grazing can be a vital tool in utilising grass during periods of challenging weather.

White clover utilisation

The advantages of legumes introduction in grassland are well documented and has been recently revised by Delaby *et al.* (2016). Caradus *et al.* (1996) outlined the major benefits associated with clover feeding, which include its effectiveness at fixing nitrogen from the air, and improving sward quality, forage intake and utilisation rates in animals.

White clover is probably the most adapted legume to improve the grass grazed system efficiency (Lüscher *et al.,* 2014). Legumes are able to fix atmospheric nitrogen, a free, natural, endless resource. The N fixation can allow for N fertilization to be reduced. This can improve the economy of the system by reducing inputs. As the N biological fixation occurs synchronously with good sward growth and is directly fixed by symbiotic nodules as organic nitrogen, the risk of negative environmental impact associated with N excess is lower. Legumes are 'unselfish' when it comes to N; it has been demonstrated than in multi-species pastures, a part of the N fixed is transferred to the grass sward.

White clover also provides advantages from an animal nutrition perspective. Research has clearly demonstrated that feeding with white clover has many benefits with respect to cow's performance due to its high nutritive value compared to perennial rye-grass. Thomson (1984) indicated that clover content in the sward needs to be at least 30%, whereas studies performed in New Zealand reported 50-60% clover content to be more appropriate to increase milk yields significantly (Harris *et al.*, 1997, 1998). Besides its high N content, white clover is a source of high quality protein for ruminants as well as minerals such as Ca. These qualities reduce the dependence of purchased protein like soybean meal or mineral supplements. White clover is characterized by a high ingestibility, high OM digestibility and excellent nutritive value. This is a consequence of the composition of the fraction of plant harvested by the animal, composed mainly of leaves and petioles. The effect of the age of regrowth on the rate of decline in nutritive value is smaller than that for the grasses. This is highly important for grazing management optimization, gives higher flexibility and permits to extend the regrowth period and compensate the negative effect of N fertilization reduction on the herbage mass (McDonagh *et al.*, 2015 – Table 4). At grazing, the advantage of PRG-WC is expressed in terms of higher intake than pure PRG at the same allowance (Ribeiro-Filho *et al.*, 2003) and in higher milk production as recently confirmed by McCarthy *et al.* (2016) (Figure 2).

These assets of white clover should not make us forget about their requirements. White clover is 'variable' and its contribution to herbage mass and persistency is difficult to predict. White clover has high demands for light and temperature. Consequently, clover growth is late to start in spring. Clover is also highly sensitive to poaching. Feeding pure white clover is not a feasible practise due to difficulties in maintaining such swards and increased bloat risk (Harris *et al.*, 1998). Bloat is a risk when clover is managed poorly, but such concerns can be overcome with better grazing supervision.

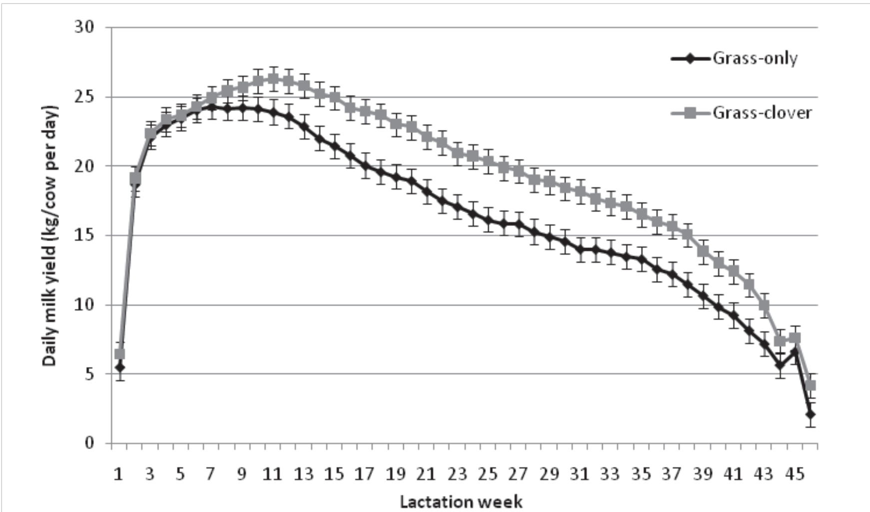

Figure 2. Daily milk production of cows grazing swards with high levels of white clover compared to cows grazing perennial ryegrass swards (McCarthy *et al.*, 2016).

Table 4. Influence of the N mineral fertilisation and the age of regrowth on herbage mass and the seasonal proportion for a pure perennial rye grass (PRG) or white clover (WC) association.

	Pure PRG		PRG + WC	
N Fertilisation (kg ha^{-1} y^{-1})	100	250	100	250
Age of regrowth (days)	30	21	30	21
Number of cycles	7	9	7	9
Herbage mass (kg DM ha^{-1})	9,125	10,555	13,020	11,450
Seasonal proportion (%) spring/summer/autumn	21/60/19	19/60/21	19/61/20	19/59/22

Cow type and welfare

The cow type required for grazing systems is different to that for indoor milk production systems. Dairy cows which are being asked to graze, must be able to maintain a compact calving pattern, have the ability to walk distances and maintain good body condition (Baumont *et al.,* 2014).

Fertility is one of the main issues as regards animal production and farm profitability across Europe. In intensively managed grass based ruminant production systems requiring seasonal calving patterns, good reproductive performance is essential (Shalloo *et al.*, 2014). Countries that are making grass based systems a priority are often adapting the genetic breeding indices to suit their systems. In 1990, the economic breeding index (Veerkamp *et al.*, 2002) was introduced to Ireland. Twenty years later, the impact can be seen with calving interval being substantially reduced in Irish herds.

Within the MultiSward project, the benefit of hybrid breeds for milk solids production and fertility was highlighted. This concept needs to be applied more in countries where grazing is to become more practised. Crossing the Holstein-Friesian breed with an alternative dairy breed sire (e.g. Normande in France, Jersey or Norwegian Red in Ireland) can provide producers with an alternative way to increase overall animal performance by increasing herd health, fertility and milk value through hybrid vigour (Delaby *et al.*, 2014; Lopez-Villalobos, 1998; Prendiville *et al.,* 2011). Suitable breeds/strains are adapted to achieving a large intake of forage relative to their potential milk yield, are fertile and healthy; have good conformation to walk long distances and high survivability (Buckley *et al.*, 2005; Dillon *et al.*, 2007). Ideally for a grass based system, dairy cows should calve early in spring every year and then immediately go to graze grass thereby resulting in the best fit between grass supply and feed demand.

Grazing has advantages with respect to animal health. Olmos *et al.* (2007) found that pasture based dairy cows had reduced lameness and better locomotive ability and also had a greater opportunity for uninterrupted lying time compared to housed dairy cows. Benefits of grass based systems in terms of lameness must be considered with caution as in certain circumstances such as when cow tracks are not maintained in grass based production systems lameness incidence can still be high. Washburn *et al.* (2002) reported that there are several examples in the literature showing that access to pasture can improve aspects of cow health such as mastitis. Incidences of environmental mastitis are low in grazing systems given that she is outdoors for long periods. The public perception of grazing allows the consumer to understand that the animal is in her natural environment and at one with nature.

New grassland technologies

The development and use of decision support tools at farm level is not a new phenomenon. The key objective of most of the tools developed is to increase the information available to help the decision making process at farm level (Minchin *et al.*, 2010). In literature there are examples of decision support tools aimed at improving the decision making process around key aspects of the farm. In the US a goal-oriented decision support tool was developed to determine the best grazing management strategies for California and other western states (Barry *et al.*, 2005). Australia has developed a range of decision support tools across different aspects of the dairy farm business (Donnelly *et al.*, 2002). Grazplan forms a family of decision support tools which are used for the maximisation of profit and environmental sustainability (Donnelly *et al.*, 2002), likewise Australia's Sustainable Grazing Systems pasture growth model also provides a management aid for livestock systems (Johnson, 2013). The advancement of the internet and in particular the proliferation of smart phones has created opportunities for the development and use of decision support tools that are web enabled (Crowley *et al.*, 2013), while facilitating the collation of a large quantities of data in a central data storage platform from different farms. The potential use of this information from a research perspective can be significant. Internationally, most decision support tools developed are stand-alone tools that do not have a data storage function which reduces the overall utility of such tools as it slows down the introduction of upgrades to the systems. The inclusion of the data storage function dramatically increases their functionality as it enables the development of longer term research based solutions established from data collected over a longitudinal time horizon across a large range of farms. It also provides an automated mechanism to benchmark farms across periods and to benchmark across a range of farms.

Efficient grazing management requires anticipation and flexibility and would be greatly facilitated by the development of dynamic tools with the capability to simulate different scenarios based on regular measurement of farm grass supply.

PastureBase Ireland

PastureBase Ireland (PBI) contains a web based PC or smart phone enabled front end and a grassland database in the back end which has the dual function of providing real time decision support for farmers and farming practitioners while capturing farm grassland data in the background for benchmarking and research purposes. PBI assists farmers, researchers and the agricultural industry to make information based grassland decisions around all aspects of grassland farming. The system operates with the individual farm paddock as the basic unit of measurement at farm level. The system is operated by the farmer entering the grassland information through the web front end. Thus, the accuracy and usefulness of the system is determined by the level and accuracy of the data recorded by the user, although it has predefined error terms programmed into the system for abnormal data. All measurements on PBI are described and calculated on a per hectare basis for individual paddocks as described below.

The operator builds a profile for each paddock by entering background paddock information such as paddock size, location on farm, altitude, aspect, soil traffic ability, paddock cultivars and soil test results which can be linked to paddock performance over time. This information is of particular interest as it can potentially assist in explaining the differences in how individual paddocks perform in comparison to others with the continuous focus to ascertain the reasons for differences in grass growth between and across paddocks. This data allows the farm to be categorised at paddock, farm and regional level for the purpose of benchmarking for both operator and research benefits. A profile of daily and cumulative grass growth for each paddock is created through the farmer entering in weekly grassland measurements. Figure 3, shows the cumulative dry matter production across Ireland dairy and drystock farms (in bold) for 2015. This shows that grazing management employed on the farm dictates the farm grass production output more than farm geographic location in Ireland

Pastur'Plan – grazing management planning

Pastur'Plan, a new tool available in France in 2015 (Delaby *et al.*, 2015), combines two concepts. The first one is adapted from the Grass Wedge (Hennessy, 2012) and the second one describes the balance

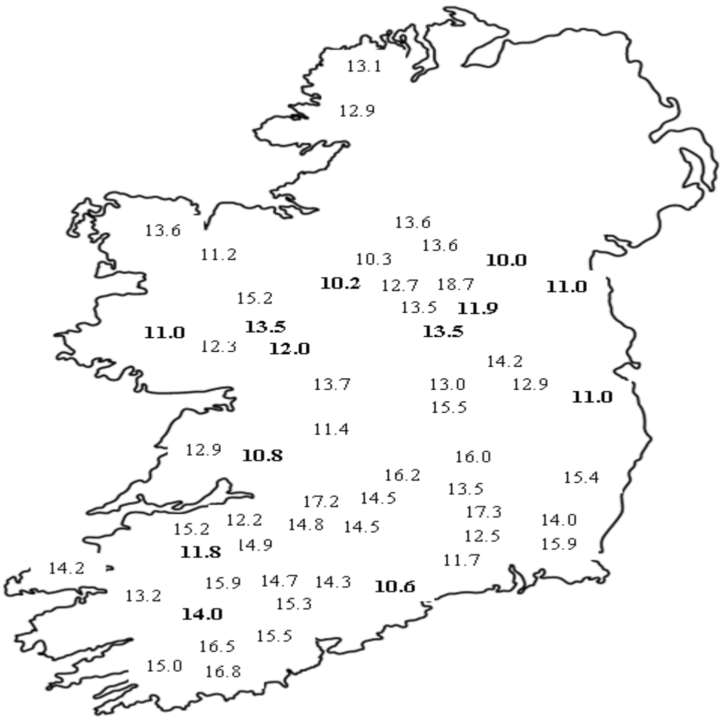

Figure 3. Regional grass production difference in Ireland in 2015 from PastureBase Ireland.

evolution between grass growth and demand according to different grazing options on a paddock by paddock basis. Taking account of (1) the herd size, the supplementation to calculate the animals grass demand and (2) the weekly grass growth profile modifiable and updatable by the user to calculate the grass available. Pastur'Plan proposes to the user (advisor or farmer) to realise the anticipated individual utilisation. This facilitates the testing of different scenarios to optimise grass utilisation. Different illustrations (Figures 4 and 5) assist in making the right decisions and propose the optimal grazing plan to the farmer. The objective of this tool is to stimulate better grazing management, improve grass utilisation and provide more confidence in grazing systems.

Other technologies

New technologies are continuously being developed and new grassland decision support tools such as the Grasshopper described by MacSweeney *et al.* (2014), cow sensors (Ipema *et al.*, 2014) and virtual fencing (MacSweeney *et al.*, 2014) will assist farmers in accurately allocating herbage to grazing dairy cows. These tools are also likely to increase farmers' confidence when it comes to grazing management and herbage allocation (Delaby *et al.*, 2015). The increase in accuracy and availability of precision technologies increases the potential integration of precision grazing into grass based systems through the replacement of some of the skills required for optimised grazing management.

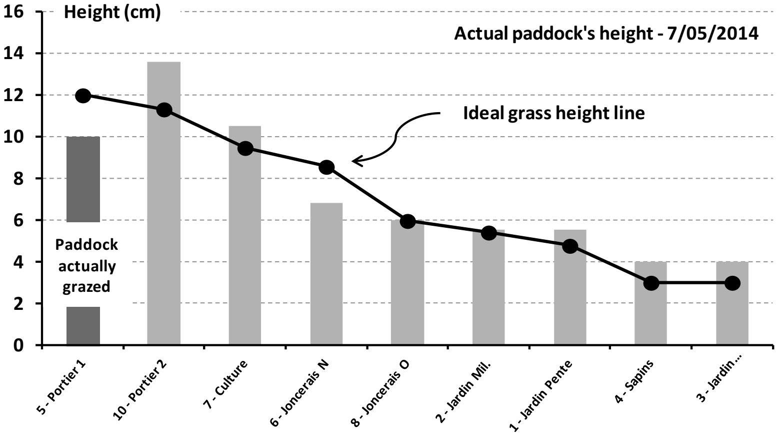

Figure 4. Example of grazing profile in Pastur'Plan.

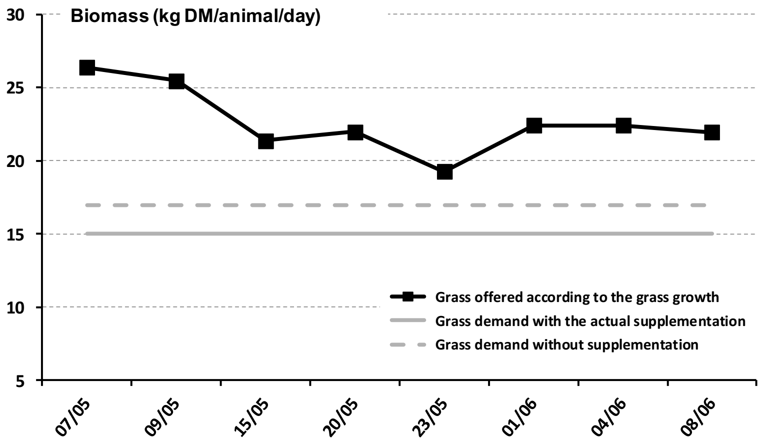

Figure 5. Grass available and grass demand evolution at each paddock change.

Conclusions

Northern Europe is well placed to make efficient use of grassland. Two reasons are now established for Europe to remerge with a better focus on grazed grass and system efficiency; i) the economic and demographic is rapidly changing in the world ii) environment sustainability can no longer be compromised. Grass based systems allow for the three pillars of sustainability to be optimised.

The mechanisms of increasing grass in the dairy cow's diet are relatively well known and will improve further with research. Grassland research results and extension knowledge is freely available in most member states, however many European farmers do not have sufficient confidence to adopt better grassland management practises to increase the proportion of grazed grass in the diet. Grassland management is challenging but many decision support tools aiming at better grassland management exist. Their use may need to be incentivised for farmers to apply them more. The emergence of focussed grassland 'discussion groups' in Ireland and France provides good opportunity to deliver more grassland theoretical knowledge, practical know-how and higher grassland ambition to farmers. It is likely that Europe's success in developing family farm based sustainable and consumer accepted dairy products will depend on its capability to use grass better in the dairy cow's diet.

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Characterisation of protein and fibre in pulp after biorefining of red clover and perennial ryegrass

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Abstract

Pressing of green forages, such as red clover (*Trifolium pratense* L.) and perennial ryegrass (*Lolium perenne* L.), result in a juice with a high content of soluble protein, and a solid pulp fraction rich in fibre and insoluble protein. The juice is – after further biorefining – a promising high quality protein source for monogastrics. The resulting pulp maintained an equal concentration of crude protein (CP) compared to the original plant, while the neutral detergent fibre (NDF) concentration doubled. The pulp dry matter (DM) concentration was above 420 g kg^{-1}, and the CP concentration was 150-210 g kg^{-1} DM. The CP proportions of the NDF fraction showed >400 g kg^{-1} of the CP content in the pulp. The chemical analyses indicate that pulp can be a promising roughage for cattle.

Keywords: protein, pulp, *Trifolium pratense*, *Lolium perenne*

Introduction

The amino acid composition of protein from green plants, harvested at early stages of maturity, is well-balanced, and extracted green protein is comparable to soya protein, and thus well suited for monogastrics (Chiesa *et al.*, 2011). A biorefining process based on screw pressing of forage produce juice rich in soluble protein and solid pulp rich in fibre and insoluble protein (Charlton *et al.*, 2009). Upon separation of the juice and pulp it was observed that the crude protein (CP) concentration in the pulp remained equal to the original plant on a dry matter (DM) basis, and the composition of amino acids in this fraction was similar to the composition in the plant (Damborg *et al.*, unpublished data). As a proportion of the protein is expected to be fibre-bound or -retained, the pulp is more suitable for ruminants than for monogastrics.

This paper highlights the chemical composition of the plants and the pulps particularly in terms of fibre and CP. Later research will focus on *in situ* nutritional evaluation. The purpose of this study was to investigate organic matter (OM) digestibility and protein content and distribution in pulp after extraction.

Materials and methods

Red clover (*Trifolium pratense* L. var. Suez) and perennial ryegrass (*Lolium perenne* L. var. Calvano 1), were grown in pure stands, under normal fertilisation practice. First regrowth and third regrowth were harvested on 13 June and 9 September 2014. The samples were taken from the windrow after mowing, frozen immediately and kept frozen until processing, where red clover (approx. 5.0 kg fresh weight) and ryegrass (approx. 3.4 kg fresh weight) were separated into juice and pulp in an Angel 8500S twin-screw press.

Fibre analyses on plant and pulp were performed according to a modified version of the Van Soest *et al.* (1991) method to determine content of neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL), and afterwards CP content in each fibre fraction. For each plant and pulp three bags (Dacron filter bags, 19 cm^2 bag size, 12 μm pore size) were filled with 10 g of sample. The filled bags were washed to eliminate immediate soluble material. All bags were then boiled in neutral detergent solution plus Na_2SO_3 and α-amylase for 1 h, and washed. One set of samples was dried for

NDF determination, and further analysis on the NDF fraction. The remaining bags were boiled in acid detergent solution and washed. One set of samples was dried for ADF determination, and further analysis on the ADF fraction. The remaining samples were immersed in 72% sulphuric acid solution for 3 h, washed and dried for ADL determination, and further analysis on the ADL fraction. Hemicellulose was determined as the difference between NDF and ADF, and cellulose was determined as the difference between ADF and ADL. DM, crude ash and nitrogen concentrations of plant and pulp samples were determined according to conventional methods. Nitrogen content was converted into CP by the factor 6.25. *In vitro* OM digestibility was performed according to Tilley *et al.* (1963).

The data analysis was generated using SAS, version 9.4 (SAS Institute Inc., Cary, NC, USA). A general linear model was applied to assess the differences between plant and pulp. Multiple comparisons were adjusted using Tukey's range test. Data is presented as means ± standard deviation. A *P*-value <0.05 was considered significant.

Results and discussion

Compared to the original plant the pulp had similar CP concentration, but lower crude ash concentration (Table 1). The *in vitro* digestibility tended to be lower for pulp, as expected, due to a large removal of soluble OM upon juice extraction. Expressed as digestible OM on DM basis (DOM), no difference was observed, due to the decrease in ash concentration between original plant and pulp.

There was a clear up-concentration of NDF and underlying constituents in pulp compared to plant. The amounts of ADL residues in perennial ryegrass were too limited to measure nitrogen content (Table 2). This corresponds well to earlier findings (Krämer *et al.*, 2012).

Table 1. Chemical composition of red clover and perennial ryegrass plant and pulp. Mean of two harvests ± standard deviation, n=2 (first and third regrowth).[1]

Plant species	Fraction	Dry matter (g kg^{-1})	Crude protein (g kg^{-1} DM)	Crude ash (g kg^{-1} DM)	*In vitro* digestibility (g kg^{-1} OM)	Digestible organic matter (g kg^{-1} DM)
Red clover	Plant	156 (±4)	213 (±23)	98 (±33)	681 (±30)	614 (±5)
	Pulp	424 (±31)	213 (±6)	72 (±37)	636 (±54)	589 (±26)
Perennial ryegrass	Plant	218 (±26)	153 (±19)	84 (±2)	722 (±19)	661 (±19)
	Pulp	456 (±48)	150 (±35)	48 (±3)	684 (±5)	652 (±3)
P-value	Fraction	<0.001	NS	0.001	0.093	NS

[1] DM = dry matter; OM = organic matter; NS = not significant.

Table 2. Concentration of NDF, hemicellulose, ADF, cellulose and ADL in red clover and perennial ryegrass plant and pulp. Mean of two harvests ± standard deviation, n=2 (first and third regrowth).[1]

Plant species	Fraction	NDF (g kg^{-1} DM)	Hemicellulose (g kg^{-1} DM)	ADF (g kg^{-1} DM)	Cellulose (g kg^{-1} DM)	ADL (g kg^{-1} DM)
Red clover	Plant	369 (±8)	131 (±5)	238 (±13)	194 (±3)	44 (±10)
	Pulp	552 (±33)	194 (±31)	358 (±3)	289 (±1)	70 (±3)
Perennial ryegrass	Plant	498 (±3)	246 (±0)	252 (±3)	239 (±2)	14 (±5)
	Pulp	706 (±46)	357 (±32)	349 (±13)	321 (±3)	28 (±17)
P-value	Fraction	<0.001	0.008	0.002	0.001	0.030

[1] ADF = acid detergent fibre; ADL = acid detergent lignin; CP = crude protein; DM = dry matter; NDF = neutral detergent fibre.

Red clover and perennial ryegrass behaved similarly upon separation, as the relative differences in OM digestibility and CP concentration between plant and pulp were uniform. The only exception was in the proportion of CP in the hemicellulose, where ryegrass showed a larger increase from plant to pulp.

The proportions of the CP in the plant or pulp samples, which was found in the fibre constituents, subsequent to Van Soest treatment showed, as expected, that a larger proportion of the CP in plant samples was lost during NDF treatment than was the case for the pulp samples, as solubles had already been partly removed by physical extraction (Table 3).

Table 3. CP proportion of the CP in the original plant or pulp sample in NDF, hemicellulose, ADF, cellulose and lignin. Mean of two harvests ± SD, n=2 (first and third regrowth).[1]

Plant species	Fraction	CP in NDF (g kg^{-1} CP in plant/pulp)	CP in hemicellulose (g kg^{-1} CP in plant/pulp)	CP in ADF (g kg^{-1} CP in plant/pulp)	CP in cellulose (g kg^{-1} CP in plant/pulp)	CP in lignin (g kg^{-1} CP in plant/pulp)
Red clover	Plant	204 (±10)	69 (±24)	134 (±14)	70 (±19)	64 (±6)
	Pulp	405 (±55)	85 (±21)	319 (±76)	184 (±73)	135 (±3)
Perennial	Plant	208 (±18)	84 (±54)	124 (±36)	–	–
ryegrass	Pulp	459 (±21)	186 (±144)	273 (±123)	239 (±141)	35 (±18)
P-value	Fraction	<0.001	NS	0.02	–	–

[1] ADF = acid detergent fibre; CP = crude protein; DM = dry matter; NDF = neutral detergent fibre; NS = not significant.

Conclusions

Pulp from both red clover and perennial ryegrass maintained CP concentrations similar to the original plant, furthermore only a non-significant decrease was seen in DOM in pulp compared to plant. These results indicate that the pulp remaining after juice extraction of green forages is an interesting roughage for ruminants.

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Upgrading of essential amino acids in plants through cattle for higher nutritional value for humans

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Abstract

Cattle production systems are characterized by great diversity. The variety of feeds and feed ration compositions is large due to ruminants' ability to digest fibrous materials inedible to humans such as roughage and by-products from the food- and biofuel industry. The aims of this study were to determine the proportions of human digestible essential amino acids (EAA) in feeds and animal products, respectively, and to compare different systems of milk and beef production, respectively. Four types of systems for milk and beef production were designed: (1) a reference similar to the typical current Swedish system; (2) intensive cattle production based on maize silage; (3) intensive forage-based production, for dairy cows complemented with food industry by-products; (4) extensive production based on forage and pasture with only small amounts of concentrate. In all four production systems the proportions of digestible EAA was generally higher in the products, milk and meat, compared to the proportions of EAA in the feeds. However, in beef production, the intensive models with dairy calves resulted in lower output in meat than input in feeds for all digestible EAA. In conclusion, except for intensive beef production with dairy calves, proportions and quality of amino acids in plants are upgraded by cattle.

Keywords: protein, roughage, by-product, dairy cow, human inedible

Introduction

Ruminants' ability to digest fibrous materials inedible to humans allows cattle production systems to be diverse and use a wide variety of feeds. In the competition of land for food or feed, marginal land and human inedible by-products are important feed resources to ruminants in order to provide food rich in protein to a growing population. Animal protein is dense in essential amino acids (EAA) which are important precursors for protein synthesis in humans and these needs to be supplied via the diet. The aim of the present study was to determine the proportions of human-digestible EAA in feeds and animal products and to compare different systems for milk and beef production.

Materials and methods

Four types of livestock production systems were designed and applied to milk and beef production: (1) a reference similar to the typical current Swedish system; (2) intensive cattle production based on maize silage; (3) intensive forage-based production, for dairy cows complemented with food industry by-products; (4) extensive production based on forage and pasture with only small amounts of concentrate. The milk systems had different annual milk yield: 9,000, 11,000 and 7,000 kg, respectively. The feed rations used in dairy production were calculated according to Swedish feeding recommendations (Spörndly, 2003). For beef, calculations were performed assuming several production models and weight gain values with finishing cattle of dairy and beef breeds on varying feed rations calculated according to NorFor (Volden, 2011). Essential amino acid concentrations and their true ileal digestibility in pigs (CVB Feed Table, 2011) were included in the calculations of input of feed and the output of EAA in milk and meat (including edible offal and blood) were calculated according to USDA (2015) and CVB Feed Table (2011). Digestible EAA input in feeds and output in milk and meat for dairy cows were calculated on annual basis. The feed consumed by the dairy replacement heifers was added to the dairy cow diet and the meat from culled cows was included in the output. For beef cattle, the input and output were calculated per slaughtered animal including input of feeds to suckler cow, breeding bull and replacement heifer and output of carcasses from dams and sires. All roughages were considered to be completely inedible to humans and the efficiency of EAA turnover was determined as the ratio of output/input.

Results and discussion

In all four milk production systems, the proportions of EAA in the products was generally higher than the proportions of EAA in the feeds and the extensive system showed the highest efficiencies (Table 1). Intensive beef production with calves originating from dairy cows resulted in a lower output in meat than input of EAA, due to the calves' need for a high-protein diet during their first six months. The milk they consume is partly produced from human-edible protein and so are the concentrates. However, by choosing a forage-based extensive rearing model, the amounts of EAA in feeds can be reduced to the same magnitude as the amounts in the meat produced for the lifetime as a whole. The extensive models of beef production with calves from suckler cows resulted in higher output in meat than input in feeds for all EAA. These calves suckle a forage-fed dam whose feed ration contains scarcely any human-edible protein. The intensive production models for beef calves generally resulted in an output of the same magnitude as the input for most EAA. Forage-based models generally resulted in higher efficiency than models based on maize silage, due to the need for extra protein feed in addition to the starch-rich, but protein-poor, maize in the latter (Table 2). In this study, we assumed that human-edible digestible EAA were absent from all forages, which biased the results towards decreased competition between human food production and animal feeds, especially in the forage-dominated systems. It can be argued that arable land in these systems is used for production of forages, when it could be used for cultivation of grain for human consumption. However, cultivation of leys is an important part of the crop rotation and may sometimes increase carbon sequestration in the soil (Soussana *et al.*, 2007). Furthermore, in areas with poor conditions for grain cropping, grasslands and forage is often not only the sole realistic alternative agricultural use of land, but also superior from a protein efficiency perspective.

Table 1. Ratio of total human-edible essential amino acids (EAA) in meat and milk: total EAA in human-edible feeds per year for different dairy production systems with different milk yields.[1]

System	Reference	Int. maize	Int. forage	Extensive
Histidine	1.13	1.35	1.06	2.15
Isoleucine	1.41	1.48	1.27	2.56
Leucine	1.54	1.63	1.32	2.77
Lysine	2.08	1.87	1.95	3.26
Methionine	1.49	2.27	1.33	2.58
Phenylalanine	1.43	1.64	1.24	2.60
Threonine	1.25	1.39	1.11	2.08
Tryptophan	1.06	1.49	0.97	2.03
Valine	1.35	1.60	1.27	2.59

[1] Reference, 9,000 kg; Intensive maize and beans, 11,000 kg; Intensive forage and food industry by-products, 11,000 kg; Extensive, 7,000 kg. The input of EAA in feeds, include cows and replacement heifers, and the output consists of EAA in milk and meat from culled cows. Values >1 indicate higher output than input of human-digestible EAA.

Table 2. Ratio of total human-edible essential amino acids (EAA) in meat: total EAA in human-edible feeds for dairy and suckler production in four systems using calves from dairy cows and beef cows.[1]

Breed	Dairy[2]					Suckler[3]						
System	Reference	Int. maize	Int. forage[4]	Extensive		Reference		Int. maize		Int. forage	Extensive	
Model[5]	Bull	Bull	Bull	Heif.	St.	Heif.	Bull	Heif.	Bull	Bull	Heif.	St.
Histidine	0.36	0.35	0.43	0.87	0.93	9.15	0.79	0.91	0.94	1.28	19.3	23.4
Isoleucine	0.30	0.31	0.37	0.75	0.80	7.78	0.67	0.80	0.81	1.09	16.4	19.9
Leucine	0.29	0.31	0.37	0.76	0.81	7.48	0.65	0.80	0.79	1.05	15.8	19.2
Lysine	0.59	0.48	0.62	1.01	1.08	16.0	1.39	1.29	1.45	2.25	33.8	41.0
Methionine	0.36	0.43	0.49	1.22	1.29	9.31	0.81	1.12	1.07	1.31	19.6	23.8
Phenylalanine	0.21	0.25	0.28	0.67	0.71	5.30	0.46	0.63	0.60	0.74	11.2	13.6
Threonine	0.30	0.31	0.38	0.78	0.83	7.91	0.69	0.80	0.82	1.11	16.7	20.2
Tryptophan	0.15	0.17	0.20	0.49	0.52	3.78	0.33	0.44	0.42	0.53	8.0	9.7
Valine	0.26	0.29	0.34	0.73	0.78	6.75	0.59	0.74	0.73	0.95	14.2	17.3

[1] Both the input of EAA in feeds and output of EAA in meat includes dam and sire in suckler production. Values >1 indicate higher output than input of human-digestible EAA.

[2] Pure dairy breed in reference, but dairy × beef cross-breeds in the other systems.

[3] Beef breed crosses.

[4] Intensive forage.

[5] Young bulls (Bull) are reared indoor, whereas heifers (Heif.) and steers (St.) are grazing during summers.

Conclusions

Forage- and pasture-based production models for milk and beef may result in upgrading of EAA compared with using the plant materials directly as human foods. This is especially important to consider in areas where grain cultivation is not an option or where conservation of grasslands is highly valued.

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Protein quality of lucerne – a comparison to red clover and effects of wilting and ensiling

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Abstract

The aim was to compare the protein quality of four lucerne varieties with the protein quality of one red clover variety and to study the effect of wilting and ensiling on the protein quality of lucerne/white clover forage. 'The Cornell Net Carbohydrate and Protein System' was used. True protein was divided into buffer-soluble protein (B1), neutral detergent (ND)-soluble protein (B2), acid-detergent (AD)-soluble protein (B3) and AD-insoluble protein (C). The remaining part of the protein was non-protein nitrogen (NPN; A). The lucerne varieties, which did not differ in protein fractions, had greater A- and B2-fractions but a smaller B3-fraction than red clover ($P<0.05$). During wilting of lucerne/white clover to 40% dry matter the B1-fraction decreased while the B3-fraction increased (P<0.001). During ensiling, the B-fractions decreased while the A-fraction increased (P<0.001). This protein degradation during ensiling was less pronounced when an acid was used as an additive, whereas a bacterial inoculant did not improve protein quality. In summary, the lucerne varieties had more NPN and rumen degradable protein but less rumen undegradable protein than the red clover variety. Wilting of lucerne/white clover improved protein quality and use of acid decreased protein degradation during ensiling.

Keywords: lucerne, white clover, wilting, ensiling, protein quality

Introduction

As lucerne (*Medicago sativa* L.) is a suitable legume in mixed legume-grass swards for silage, it is important to investigate potential differences in protein quality between some typical lucerne varieties and to relate the protein quality of lucerne with that of red clover (*Trifolium pratense* L.). Wilting is beneficial as dry matter (DM) concentration of lucerne is negatively correlated to proteolysis during ensiling (Muck *et al.*, 2003). Additives can further restrict proteolysis of lucerne silage (Nadeau *et al.*, 2000). The objective of this study was to compare the protein quality of four lucerne varieties and to compare those with the protein quality of one red clover variety. Furthermore, effects of wilting, ensiling and additive on the protein quality of lucerne/white clover forage were evaluated.

Materials and methods

The four lucerne varieties Pondus (SW, Sweden), Genoa (Syngenta, USA), Daphne (Florimond Desprez group) and Creno (DLF-Trifolium) and one red clover variety (Titus, 4n, Saatzucht Steinach, GmbH) were grown in a randomized block design with three field blocks at the Rural Economy and Agricultural Society Sjuhärad, southwest Sweden (57°36′N, 13°15′E). Forages were harvested as spring growth and as first and second regrowth on June 15, July 26 and September 4, 2012, respectively. Forage maturity was early bud to bud in lucerne and late bud in red clover in the spring growth and early bloom in the regrowths for all the species except Creno, which was in the bud stage. Forages were sorted from white clover and weeds and analysed for crude protein (CP) fractions according to Licitra *et al.* (1996). True protein (TP) was divided into buffer-soluble protein (BSP; B1, rapid rumen degradation), neutral detergent (ND)-soluble protein (NDSP; B2, variable rumen degradability), acid-detergent (AD)-soluble protein (ADSP; B3, slowly degraded, much escapes rumen degradation) and AD-insoluble protein

(ADIP; C, indigestible). The remaining part of the CP was non-protein nitrogen (NPN; A, some instantly used as ammonia, some lost as urea in urine; Sniffen *et al.,* 1992). During the first regrowth, a mixture of the four lucerne varieties, including 15% white clover (*Trifolium repens* L.) and some weeds, was chopped and wilted for 6 h to 40% DM before being ensiled in 1.7 l silos for 90 d. Lucerne/white clover was either untreated or treated with Kofasil Lac (*Lactobacillus plantarum* DSM 3676, 3677; Addcon Europe GmbH) at 100,000 cfu g^{-1} or GrasAAT SP (formic acid, propionic acid, sodium formate, sodium benzoate; Addcon Nordic AS) at 4 l T^{-1}. Data for fresh forage of the four lucerne varieties and the red clover variety were analysed by harvest. Data from the silage experiment were analysed as a completely randomized design using three replicates per treatment.

Results and discussion

CP concentration was similar between lucerne and red clover in the spring growth but it was generally greater in lucerne than in red clover in the regrowths (Table 1). TP concentration was smaller in lucerne

Table 1. Crude protein (CP), true protein (TP) and CP fractions of red clover and lucerne varieties in spring growth and first and second regrowth.[1]

	Red clover Titus	Lucerne					
		Pondus	Genoa	Daphne	Creno	SEM	*P*-value
Spring growth							
CP, g kg^{-1} DM	186	180	189	190	203	5.2	ns
TP, g kg^{-1} DM	133^{a}	104^{b}	111^{b}	107^{b}	119^{a,b}	4.8	<0.05
g kg^{-1} CP[1]							
NPN (A)	283^{b}	422^{a}	413^{a}	436^{a}	413^{a}	20.4	0.01
BSP (B1)	17	18	24	16	24	6.2	ns
NDSP (B2)	305^{b}	431^{a}	429^{a}	421^{a}	441^{a}	25.4	<0.05
ADSP (B3)	337^{a}	77^{b}	88^{b}	80^{b}	81^{b}	15.2	<0.001
ADIP (C)	58^{a}	52^{a,b}	47^{b,c}	48^{b,c}	41^{c}	0.29	<0.05
First regrowth							
CP, g kg^{-1} DM	141^{b}	212^{a}	211^{a}	206^{a}	201^{a}	7.6	<0.001
TP, g kg^{-1} DM	111^{c}	115^{b,c}	122^{a}	119^{a,b}	124^{a}	1.9	<0.01
g kg^{-1} CP[1]							
NPN (A)	211^{b}	455^{a}	423^{a}	423^{a}	384^{a}	24.4	<0.001
BSP (B1)	28	27	20	15	15	4.8	ns
NDSP (B2)	239^{b}	390^{a}	415^{a}	413^{a}	440^{a}	16.8	<0.001
ADSP (B3)	457^{a}	87^{b}	92^{b}	107^{b}	109^{b}	11.4	<0.001
ADIP (C)	66^{a}	41^{b}	50^{a,b}	42^{b}	52^{a,b}	5.0	<0.05
Second regrowth							
CP, g kg^{-1} DM	221^{c}	236^{b,c}	248^{a,b}	250^{a,b}	254^{a}	5.0	<0.01
TP, g kg^{-1} DM	164^{a}	138^{b}	145^{b}	136^{b}	141^{b}	4.1	<0.01
g kg^{-1} CP[1]							
NPN (A)	256^{b}	412^{a}	415^{a}	454^{a}	445^{a}	19.3	<0.001
BSP (B1)	17	33	27	5	31	8.4	ns
NDSP (B2)	267^{c}	434^{a}	430^{a}	414^{a,b}	385^{b}	10.3	<0.001
ADSP (B3)	408^{a}	75^{b}	84^{b}	65^{b}	90^{b}	13.4	<0.001
ADIP (C)	52^{a}	46^{a,b,c}	42^{b,c}	42^{c}	49^{a,b}	2.2	<0.05

[1] BSP = buffer-soluble protein; ADSP = acid-detergent soluble protein; ADIP = acid detergent insoluble protein; NDSP = neutral detergent soluble protein; NPN = non-protein nitrogen; SEM = standard error of the mean; ns = not significant, P>0.10.

Table 2. Crude protein (CP), true protein (TP) and CP fractions of lucerne/white clover forage as affected by wilting and ensiling (90 d of storage).[1]

	Unwilted forage	Wilted forage	Untreated silage	SEM	*P*-value
CP, g kg^{-1} DM	190^{b}	200^{a}	204^{a}	1.8	<0.01
TP, g kg^{-1} DM	140^{a}	144^{a}	79^{b}	1.5	<0.001
NPN (A), g kg^{-1}CP	262^{b}	283^{b}	612^{a}	10.3	<0.001
BSP (B1), g kg^{-1}CP	169^{a}	74^{b}	27^{c}	8.8	<0.001
NDSP (B2), g kg^{-1}CP	494^{a}	513^{a}	270^{b}	6.6	<0.001
ADSP (B3), g kg^{-1}CP	26^{c}	72^{a}	38^{b}	0.86	<0.001
ADIP (C), g kg^{-1}CP	49	59	53	2.7	ns

[1] BSP =buffer-soluble protein; ADSP = acid-detergent soluble protein; ADIP = acid detergent insoluble protein; NDSP = neutral detergent soluble protein; NPN = non-protein nitrogen; SEM = standard error of the mean; ns = not significant, *P*>0.10.

Table 3. Crude protein (CP), true protein (TP) and CP fractions of untreated and treated lucerne/white clover silage after 90 d of storage.[1]

	Untreated silage	Kofasil Lac	GrasAAT SP	SEM	*P*-value
CP, g kg^{-1} DM	204	200	198	3.7	ns
TP, g kg^{-1} DM	$79^{(b)}$	$74^{(b)}$	$88^{(a)}$	3.3	0.060
NPN (A), g kg^{-1}CP	612^{a}	632^{a}	554^{b}	14.9	<0.05
BSP (B1), g kg^{-1}CP	27	21	22	10.2	ns
NDSP (B2), g kg^{-1}CP	$270^{(b)}$	$273^{(b)}$	$327^{(a)}$	17.1	0.095
ADSP (B3), g kg^{-1}CP	38^{a}	23^{b}	42^{a}	3.5	<0.05
ADIP (C), g kg^{-1}CP	53	51	54	1.98	ns

[1] BSP =buffer-soluble protein; ADSP = acid-detergent soluble protein; ADIP = acid detergent insoluble protein; NDSP = neutral detergent soluble protein; NPN = non-protein nitrogen; SEM = standard error of the mean; ns = not significant, *P*>0.10.

than in red clover in the spring growth and in the second regrowth but generally greater than in red clover in the first regrowth although the differences were small.

There were usually no differences in CP and TP concentrations between the lucerne varieties. NPN and NDSP fractions were larger whereas ADSP was smaller in lucerne than in red clover in all harvests. There were only small or no differences in ADIP between lucerne and red clover. Generally, CP fractions were similar between the lucerne varieties (Table 1). Thus, red clover contains more protein that escapes rumen degradation and is absorbed in the duodenum (Sniffen *et al.,* 1992).

During wilting of lucerne/white clover forage the BSP decreased and the more nutritionally favoured ADSP increased, with no increase in ADIP (Table 2). During ensiling, the B fractions of the TP decreased while the NPN increased (Table 3). GrasAAT SP decreased proteolysis during ensiling as shown by more NDSP and less NPN compared to the untreated silage, which improves protein utilisation by ruminants (Sniffen *et al.,* 1992).

Conclusions

Lucerne contained more NPN and more protein that is degraded in the rumen compared to red clover. Wilting increased the slowly degraded protein and rumen undegraded protein of lucerne/white clover by increasing ADSP. Proteolysis during ensiling was restricted by use of GrasAAT SP.

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Comparison of pasture based feeding systems and a total mixed ration feeding system on dairy cow milk production

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Abstract

Feeding system can have an impact on milk yield and milk solids yield. It is well recognised that cows fed a total mixed ration (TMR) have higher milk production than those predominantly fed grazed pasture for the entire lactation. Research indicates that incorporating white clover (*Trifolium repens* L.) into grass swards can have a positive impact on milk production. The objective of this experiment was to examine the effect of feeding system on dairy cow milk yield and milk solids (MS) yield. The experiment had three treatments: (1) TMR (grass silage, maize silage, concentrate); (2) grazed grass only receiving 250 kg N ha^{-1} (GO); and (3) grazed grass-white clover receiving 250 kg N ha^{-1} (GC). Total concentrate fed per lactation was 285 kg cow^{-1} for GO and GC, and 2,910 kg cow^{-1} for TMR. The GO treatment (15,909 kg DM ha^{-1}) had greater (P<0.05) cumulative herbage production than GC (14,122 kg DM ha^{-1}). There was a treatment effect (P<0.05) on milk yield and MS yield per cow whereby the TMR treatment had the highest cumulative milk and MS yield (7,815 kg cow^{-1} and 632 kg cow^{-1}, respectively). This was greater than the GC treatment (6,881 kg milk yield and 554 kg MS) and the GO treatment (6,140 kg milk yield and 515 kg MS). Incorporating white clover into the grass sward increased milk production per cow (+741 kg milk yield and +39 kg MS cow^{-1}) compared to grass only.

Keywords: white clover, grass, total mixed ration, milk production

Introduction

Apart from an animal's genetic capabilities, feed quality is the single biggest determinant of milk production in any given milk production system. In pasture-based systems the quality of the diet is variable and milk production per cow is often lower than when feeding total mixed ration (TMR) diets (Bargo *et al.,* 2002; Kolver and Muller, 1998). These authors suggest that, in grazing conditions, energy intake is the most limiting factor for milk production (Bargo *et al.,* 2003; Kolver and Muller, 1998), and a supplementation strategy is required to obtain higher milk production. There is renewed interest in incorporating white clover (*Trifolium repens* L.; clover) into grass based milk production systems in Ireland due to its ability to fix nitrogen, improve pasture quality and utilisation, and increase milk production (Enriquez-Hidalgo *et al.,* in press). The objective of this experiment was to examine the effect of feeding system on dairy cow milk yield and milk solids (MS) yield by comparing three feeding systems: total mixed ration (TMR), grazed grass only (GO) and grazed grass-clover (GC).

Materials and methods

A full lactation farm systems experiment (February to November) was undertaken at Teagasc, Animal and Grassland Research Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland in 2015. The experiment had three treatments: (1) TMR (grass silage, maize silage, concentrate); (2) grass only receiving 250 kg N ha^{-1} (GO); and (3) grass-clover receiving 250 kg N ha^{-1} (GC). The TMR diet consisted of, on a DM basis, 7.15 kg grass silage, 7.15 kg maize silage and 8.3 kg concentrates. Average annual sward clover content of the GC treatment was 24%. The GO and GC treatments received 285 kg of concentrate cow^{-1} lactation^{-1}, fed in February, March and November, and the TMR treatment received 2,910 kg concentrate cow^{-1} lactation^{-1}. The grazing treatments (GO and GC) were stocked at 2.73 cows ha^{-1}

in a closed farm system (i.e. the farmlet provides all grazed herbage and silage for the animals in the treatment). Both grazing groups were rotationally grazed, achieving 8.3 rotations in the season. Fifty-four spring calving Friesian and Friesian × Jersey dairy cows were blocked on calving date, breed, pre-experimental milk yield (MY) (milk yield for first 2 weeks of lactation), pre-experimental milk solids (MS) yield (as for MY), and parity, and randomly allocated to one of the three treatments (n=18). Cows remained in their treatment groups for the entire lactation. Swards were rotationally grazed with herbage allocated to the grazing groups each day to achieve a post grazing sward height of 4 cm. Pasture allocation for the grazing treatments was determined using pre-grazing herbage mass (>4 cm) and area (m^2). Pre-grazing herbage mass was measured with an Etesia mower (Etesia UK Ltd., Warwick, UK) twice weekly. Pre- and post-grazing sward heights were measured every day using the rising plate meter (Jenquip, Fielding, New Zealand). Sward clover content was measured in each GC paddock prior to grazing as described by Egan *et al.* (2013). Cows on the TMR treatment were housed on cubicles with rubber mats and fed at 08:30 h each day. Cows were fed *ad libitum* to approximately 10% refusal levels. The feed was distributed into electronically controlled Griffith Elder Mealmaster individual feed bins (Griffith Elder and Company Ltd, Suffolk, UK) which allowed the feeding habits and daily intake of each cow to be monitored. Samples of the individual TMR components were taken weekly to determine DM, crude protein, fibre and energy contents, and diets adjusted accordingly to maintain a constant crude protein and energy supply. Milk yield was measured daily and milk composition (fat, protein, lactose) was measured weekly. Milking took place at 07:30 and 15:30 h daily. Individual milk yields (kg) were recorded at each milking using DairyMaster milk meters (Dairymaster, Causeway, Co. Kerry, Ireland). Milk fat and protein concentrations were calculated weekly from one successive evening (Monday) and morning (Tuesday) milking for each animal using a MilkoScan 7000 (Foss Ireland Ltd.). Data were analysed using PROC MIXED in SAS (SAS Institute, Cary, NC, USA) where the fixed terms were treatment and lactation, the random term was cow, and week was a repeated measure.

Results and discussion

There was a significant difference (P<0.05) in cumulative herbage production between the two grazing treatments. The GO and GC treatments grew 15,909 and 14,122 kg DM ha^{-1}, respectively. The TMR treatment had greater (P<0.05) cumulative milk yield and MS yield than the GO and GC treatments (Table 1). Though not statistically significant (P=0.1217), clover inclusion in the diet increased MS yield by 39 kg MS cow^{-1} or 106.5 kg MS ha^{-1} compared to the GO treatment. This increase in MS yield from the GC treatment can be attributed to an increase in milk yield rather than from increased milk constituents (Table 1). The GO treatment produced milk with greater (P<0.05) fat and protein concentrations than the GC and TMR treatments (Table 1). There was a significant treatment × week interaction effect on MS yield. The TMR treatment had higher (P<0.05) weekly MS yield that the GO

Table 1. Daily and cumulative milk production of cows grazing grass only receiving 250 kg N ha^{-1} + 285 kg concentrate cow^{-1} lactation^{-1} (GO), cows grazing grass clover receiving 250 kg N ha^{-1} + 285 kg concentrate cow^{-1} lactation^{-1} (GC) and cows fed on a total mixed ration diet indoors (TMR; 2910 kg concentrate cow^{-1} lactation^{-1}) over a full lactation (February to November 2015).[1]

	GO	GC	TMR	SE	TRT
Milk yield (kg cow^{-1} day^{-1})	21.12	22.29	25.74	1.318	<0.05
Milk solids yield (kg cow^{-1} day^{-1})	1.85	1.93	2.23	0.063	<0.0001
Milk fat content (g kg^{-1})	50.0	47.5	47.8	0.100	NS
Milk protein (g kg^{-1})	37.4	36.3	35.0	0.060	<0.05
Cumulative milk yield (kg cow^{-1})	6140	6881	7815	219.436	<0.0001
Cumulative milk solids yield (kg cow^{-1})	515	554	632	15.345	<0.0001

[1] SE = standard error; NS = not significant; TRT = treatment.

and GC treatments. Weekly MS yield was different ($P<0.001$) amongst treatments from week nine of the experiment until the end of lactation (Figure 1). The TMR treatment had greater milk production than the GO and GC treatments, most likely due to the higher and more consistent quality of the TMR diet compared to the pasture based diets (Bargo *et al.,* 2002; Kolver and Muller, 1998). Similar to Egan *et al.* (2015), the GC treatment resulted in increased milk production compared to the GO, particularly from June until the end of lactation when sward clover content increases resulting in increased herbage quality.

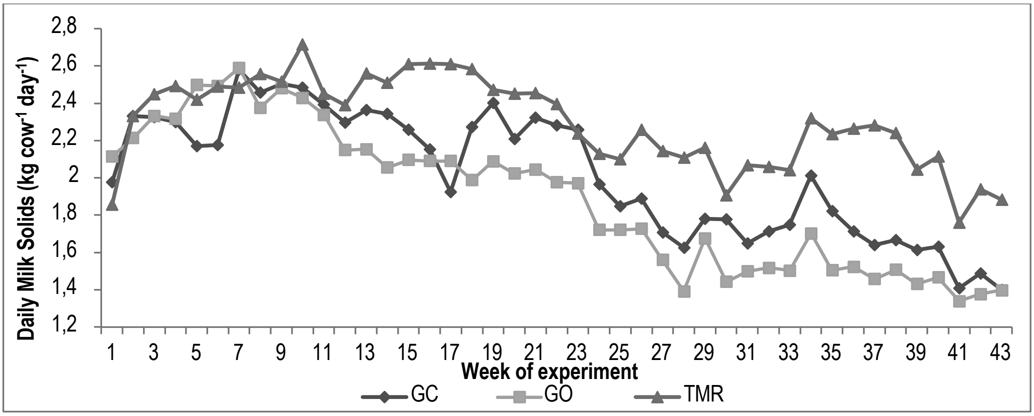

Figure 1. Daily milk solids yield (kg MS cow^{-1} day^{-1}) of cows grazing grass only receiving 250 kg N ha^{-1} + 285 kg concentrate cow^{-1} lactation^{-1} (GO), cows grazing grass clover receiving 250 kg N ha^{-1} + 285 kg concentrate cow^{-1} lactation^{-1} (GC) and cows fed on a total mixed ration diet indoors (TMR; 2910 kg concentrate cow^{-1} lactation^{-1}) over a full lactation (February to November 2015).

Conclusions

Clover inclusion did not increase total sward herbage production. Feed system had a significant effect on milk yield and milks solids yield with the TMR treatment having greater milk production than both of the grazing groups.

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Milk production potential of regrowth grass silages

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Abstract

The aim of the study was to evaluate the production potential of regrowth-grass silages. It was hypothesized that milk production is smaller than expected with highly digestible third-cut silage when compared with second-cut silages. The experiment was conducted using 39 dairy cows. Three different regrowth-silages were compared: early second-cut (10.4 MJ metabolisable energy (ME) kg^{-1} dry matter (DM)), late second-cut (9.8 MJ ME kg^{-1} DM) and third-cut (10.9 MJ ME kg^{-1} DM). Diets were fed as total mixed ration with a target concentrate proportion of 450 g kg^{-1}. Concentrate contained barley (800 g kg^{-1}) and rapeseed meal (200 g kg^{-1}). Third-cut silage had the lowest DM intake and late second-cut silage was the highest (10.7 and 11.7 kg d^{-1}). This compensated for differences in silage energy content, thus resulting in relatively small differences in ME intake between the treatments. The lowest feed efficiency and milk yield (29.1 kg d^{-1}) occurred with late second-cut, as expected. However, the difference in milk yield was only 1.4 kg between late second-cut and third-cut, despite large difference in silage energy content. There was no significant difference in milk yield between early second-cut (30.7 kg d^{-1}) and third-cut silage (30.5 kg d^{-1}). This study provides evidence for overestimation of milk production potential of autumn-harvested third-cut silage.

Keywords: harvest timing, milk yield, grass silage

Introduction

The digestibility of grass can greatly affect the milk production of dairy cows. The organic matter digestibility of early harvested grass is usually high. Grass harvested at the third cut can also have high digestibility as a consequence of low amounts of fibre in northern climate zones; this is due to low average temperatures and low radiation during the late summer period (Thorvaldsson *et al.*, 2007). Experiments using third-cut silage have shown variable results for milk production potential, but in some cases third-cut silage has resulted in lower milk production than expected (Huhtanen *et al.*, 2001; Sairanen and Juutinen, 2013).

The metabolisable energy content of grass silages is calculated by multiplying the content of digestible organic matter (g kg^{-1} dry matter (DM); D-value) with a coefficient of 0.016. This coefficient is the same for all harvests. However, it has been hypothesized that highly digestible grass contains components, e.g. phenolic compounds that might increase losses of methane and urine energy. Thus third-cut silage might have lower efficiency of metabolisable energy (ME) utilization for milk (k_l; Bruinenberg *et al.*, 2002) and the coefficient for silage ME content should be smaller than 0.016.

The objective of this study was to evaluate the feeding value of third-cut grass silage compared with that of second-cut silage.

Materials and methods

Silage crops were harvested consecutively from the same timothy-meadow fescue sward in 2014. The silages were prepared on 24 July (early second-cut, 10.4 MJ ME kg^{-1} DM), 6 August (late second-cut, 9.8 MJ ME kg^{-1} DM) and 1 September (third-cut, 10.9 MJ ME kg^{-1} DM). Diets were fed as total mixed ration (TMR) with a target concentrate percentage of 450 g kg^{-1}. The concentrate contained barley at

800 g kg^{-1} and rapeseed meal at 200 g kg^{-1} on an air-dry basis. The experiment was conducted using 39 Holstein and Nordic Red dairy cows. The average milk yield at the beginning of the experiment was 33.8 kg per day and the cows were, on average, 115 days in milk. The experiment had an incomplete cross-over design with two periods. The transition time between the periods was 17 d and data collection lasted for 7 d. The organic matter digestibility of the silages was based on pepsin-cellulase solubility.

Results and discussion

The DM intake was the lowest with the third-cut silage. Due to TMR feeding the concentrate intake was 0.5 kg DM lower in the third-cut silage treatment compared with the other treatments (Table 1). The DM contents of the experimental TMR were adjusted to be similar by additional water so that differences in TMR dry matter content did not affect the results. The fermentation quality was comparable or better with third-cut silage, compared with early second-cut silage. The silage dry matter intake index (SDMI; Huhtanen *et al.,* 2007) was the same with third-cut silage and late second-cut silage (98). The lowest SDMI was with the early second-cut silage (95). Thus the chemical properties did not explain the lower intake in third-cut silage compared with other experimental diets. Weather conditions are variable during autumn and for this reason the herbage could contain variable microbial flora or be affected by diseases that result in decreased intake of the silage. These types of quality parameters do not necessarily affect chemical analysis and cause variation in experimental results.

The lowest proportion of silage neutral detergent fibre (NDF) in the total DM intake was 270 g kg^{-1} which was with the third-cut silage. Finnish diet formulation uses the lower limit of 250 g kg^{-1} silage NDF as a physiologically recommended diet. Thus the NDF content in third-cut silage was adequate for inclusion in feed rations.

The late second-cut harvested silage resulted in the lowest milk yield, whereas there was no significant difference in milk production between early-second and third-cut silages. The difference in milk production (0.6 kg energy corrected milk (ECM) 10^{-1} g silage D-value) between early and late-second cuts was the same as reported by Kuoppala *et al.* (2008). Third-cut silage tended to produce a lower amount of ECM compared with early second-cut silage. The milk production results are in agreement with findings reported by Sairanen and Juutinen (2013) and Huhtanen *et al.* (2001). The calculated

Table 1. Intake, milk production and energy utilization with three experimental silages.[1,2]

	Early-second	Late-second	Third-cut	SEM	*P*-value
Intake, kg DM d-1					
Silage	11.4 a	11.7 a	10.7 b	0.14	<0.001
Concentrate	9.0 a	9.0 a	8.5 b	0.13	0.005
Total	20.4 a	20.7 a	19.3 b	0.25	<0.001
ME, MJ ME d^{-1}	234 a	230 ab	227 b	2.94	0.040
Milk production, kg d-1					
Milk	30.7 a	29.1 b	30.5 a	0.4	<0.001
ECM	31.9 a	29.6 b	31.2 a	0.39	<0.001
Feed efficiency					
k_l	0.59 a	0.56 b	0.61 a	0.008	0.019
kg ECM kg^{-1} DM	1.43 a	1.56 b	1.61 c	0.020	0.010
kg milk kg^{-1} DM	1.40 a	1.50 b	1.57 c	0.021	<0.001

[1] DM = dry matter; ME = metabolisable energy; ECM = energy corrected milk; SEM = standard error of the mean.

[2] Values within a row with a different letter are significantly different.

ECM yield for third-cut silage would have been 33.8 kg based on a response of 0.6 kg ECM per 10 g increase in silage D-value.

The milk production did not respond to increased silage D-value between second-cut and third-cut silage as expected based on differences in D-values. The reason was not energy losses via methane or urine, because the feed efficiency was the highest in third-cut silage. Energy utilization in milk production, k_l, was at the same level with the early-second cut and the third-cut silage. Intake was limited in third-cut silage for reasons that are unknown. Decreased intake combined with unchanged milk production with third-cut silage led to a decreased energy balance. Low energy balance is typically linked with improved apparent energy utilization. It is possible that low intake of third-cut silage was compensated by tissue mobilization, although changes in live weight are not reliable in short-term experiments.

Conclusions

Third-cut silage was higher in energy content compared with second-cut silages. The high energy content was not realized as milk production. Low total feed DM intake was the reason for the relatively low milk yield with third-cut silage. This study did not support the hypothesis of increased metabolic losses with high digestibility of third-cut compared with second-cut silages.

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The economics of grass and red clover silage yield and quality in organic dairy system

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Abstract

The effects of cutting frequency, silage fermentation pattern and legume performance in grass-clover ley on use of inputs and profitability in an organic dairy system in Mid-Norway were examined. A whole-farm linear programming model was developed to compare a three-cut and a two-cut system, either with restricted silage fermentation through acidification or untreated at low or high red clover (*Trifolium pratense* L) proportion in the ley. Input-output relations incorporated into the model were derived from a meta-analysis of organic grassland field trials in Norway, silage fermentation experiments, and with feed intakes and milk yields from simulations with the NorFor feed evaluation system. The model maximised total gross margin of farms with 250,000 l milk quota, and housing capacity for 45 cows. Farmland availability was allowed to vary with 40 ha as the basis. A high proportion of legumes in the leys was far more important for profitability than the score on the other variables considered. With little land available, the costs of preservatives were higher than their benefits. At higher land areas, applying preservatives was more profitable. Cutting systems producing silages that result in an increased intake of silage per cow, generally three-cut systems, performed relatively better at higher land availabilities.

Keywords: digestibility, *Trifolium pratense*, preservatives, milk response, land availability, linear programming

Introduction

Forage legumes, normally as components of grass-clover leys, contribute to biological nitrogen fixation, and are crucial for forage yield and profits in organic dairy systems. Milk production is, in addition to forage availability, also highly dependent on the forage feed quality. Forage feed quality is governed by the maturity of the crop at harvests. Farmland availability profoundly influences the profitability of grass silages harvested at early maturity stages in conventional dairy systems (Flaten *et al.*, 2015). Few studies have however examined the economics of different harvesting regimes in organic dairying, which has lower forage output and reduced concentrate feeding compared to non-organic systems. Moreover, the clover proportion and protein concentration differ between cuts, and are usually much higher in the regrowths than in the spring growth (Steinshamn *et al.*, 2016). Fermentation of silage further influences feed value of forage by reducing voluntary intake and utilisation of digestible nutrients. Preservatives control and direct silage fermentation, but it is unknown if the benefits of preservatives that improve animal production responses offset the costs.

Clearly, there is a need for more knowledge of optimal forage quality strategies under organic dairy management. This paper examines how cutting frequency, silage fermentation pattern and legume performance in grass-clover ley influence use of inputs and profitability in an organic dairy system at varying farmland availability.

Materials and methods

A whole-farm linear programming model of an organic dairy farm was developed to compare a three-cut and a two-cut system, either with restricted fermentation through acidification or untreated at low or high red clover proportion in the ley (8 versions of the model in all). The model includes common activities and constraints to organic dairy farms in Norway. Important activities are: (1) crop production; land can be used for growing grass/clover (for pasture or silage making) or barley, and with various levels of manure applications to some of the crop activities distinguished; (2) purchase of a variety of concentrates with different protein levels, (3) livestock production with dairy cows (replacement heifers are assumed purchased), (4) purchase, sale and application of manure, (5) field operations such as harvesting of grain and grass and silage making of grass in round bales, and (6) government payments. Constraints of the model include fixed resources of the farm (e.g. land, labour, milk quota, and housing requirements), links between activities, and organic legislation.

Grass yields for silage production were derived from a meta-analysis of organic grassland field trials at Kvithamar Research Station in Mid-Norway (Steinshamn *et al.*, 2016). We examined two-cut and three-cut systems, both cutting systems with a low (10%) and a high (40%) clover proportion of the annual yield. Dry matter (DM) grass yields were in general 12% higher, but with lower digestibility, in the two-cut than in the three-cut systems. DM yields of high-clover leys were 20-30% higher than low-clover leys. Silage fermentation experiments showed that formic-acid preservatives restricted the production of organic acids and preserved proteins compared to untreated silage (Bakken *et al.*, 2014).

The Norwegian version (TINE Optifor) of the non-additive Nordic feed evaluation system Norfor (Volden, 2011) was used to find the rations with lowest costs at various feed energy levels. The proportion of first cut silage in the diets was constant throughout the year. The Norfor system assumes a linear milk response to changes in dietary nutrient supply. Jensen *et al.* (2015) found, however, that multiparous dairy cows had higher and more nonlinear responses in milk production to increased energy intake compared to primiparous cows. We used data from Jensen *et al.* (2015) to adjust milk production responses, and separated the dietary requirements and milk responses of the cows in three age classes; first calvers, second calvers, and older. The replacement rate was 40%.

The model maximises total gross margin, that is, returns from livestock and cash crop production and government payments minus variable costs of production (2014-prices; € 1 ≈ NOK 8.40; NOK = Norwegian krones). The farm has a milk quota of 250,000 l, and housing capacity for 45 cows. Farmland availability was allowed to vary with 40 ha as the basis.

Results and discussion

At 40 ha, high legume leys resulted in much higher total silage DM supplies, more cows and higher milk production than low legume leys (Table 1). Total gross margins for high legume systems were NOK 40,000-70,000 higher than for comparable low legume systems – at 40 ha as well as for most comparisons at 20 or 70 ha. These findings confirm the central role a successful legume performance plays for production and profitability in organic systems.

The three-cut systems supplied less silage DM than the two-cut systems. At 40 ha, the number of cows and milk sold were usually highest for the two-cut systems. Profitability was however increased for most three-cut systems compared to two-cut systems at 40 ha, since higher digestibility of silage from three-cut system improved animal performance. Higher farmland availability tended to increase the profitability of highly digestible silage, as reported in studies of non-organic dairy systems (e.g. Flaten *et al.*, 2015). The comparison of the low clover natural fermentation (LCNF) systems, however, showed the opposite trend because, in contrast to the other cutting comparisons, forage intake per cow with LCNF was highest for two-cut silage.

Application of preservatives was only profitable in some of the natural fermentation/restricted fermentation comparisons at 70 ha. Enough silage was then available on the farm to take advantage of the

Table 1. Annual production results for the eight model versions[1] at 40 ha land available[2], 250,000 l milk quota and 45 cow places. Financial results at 20, 40, and 70 ha of land.

	2LCNF	2LCRF	2HCNF	2HCRF	3LCNF	3LCRF	3HCNF	3HCRF
Production results (40 ha)								
Number of cows	29.7	29.7	40.1	37.3	30.5	27.8	36.3	34.1
Milk sold (1000 l)	169	169	228	212	181	160	213	200
Concentrates (t DM)	32	32	66	44	45	22	62	48
Total silage supply (t DM)	125	125	156	159	121	123	142	144
Purchase of manure (t)	0	0	0	-187	125	-66	0	52
Total gross margins (1000 NOK)								
20 ha land	498	485	552	541	503	493	547	531
40 ha land	799	774	857	847	805	806	865	842
70 ha land	1,178	1,141	1,179	1,207	1,164	1,198	1,204	1,203

[1] Acronyms: 2/3: 2 or 3 cuts; LC/HC: low or high clover proportion; NF/RF: natural or restricted fermentation; DM: dry matter.

[2] At 40 ha: Only grass-clover or grass-clover undersown in barley in the establishment year. Grain grown singly is introduced at higher land availabilities.

positive effect on feed intake by a lower content of fermentation products and better protein preservation in additive-treated silages. At lower areas, costs of applying preservatives exceeded the benefits. It should be kept in mind that these findings are based on mathematical modelling of animal processes via the Norfor system rather than observed animal performances, e.g. by experimentation, and we assumed similar storage and feed-out losses across the different alternatives.

Conclusions

The most striking feature of this study was the great importance of legumes for high forage yields and profitability in the organic dairy system examined. Cutting systems producing silages that result in an increased intake of silage per cow, generally three-cut systems, performed relatively better at higher land availabilities. The profitability of applying preservatives to silages was questionable, in particular under restricted land availability.

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Effects of silage additives and aerobic exposure before feeding on feed intake and growth of ewe lambs

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Abstract

Fermentation quality of silage depends on the ensiling method, while hygienic quality is affected by microbial growth in the silage after given access to air. This study used 24 weaned ewe lambs to investigate how feed intake and growth was affected by silage additive treatment, aerobic storage time of silage before feeding, and concentrate level. Heated or moulded silage was not offered to animals, and not included in feed samples. Acidic silage additive improved silage quality and resulted in improved silage intake (0.07 kg DM day^{-1}) and weight gain (39 g day^{-1}) on average for silage not exposed, or exposed to air before feeding. There was no main effect of aerobic storage time of silage before feeding. Increased concentrate level from 100 to 300 g day^{-1} increased weight gain of the lambs by 29 g day^{-1}.

Keywords: silage, silage additives, aerobic exposure, feed intake, growth, ewe lambs

Introduction

Sheep farming in Norway is mainly a roughage-based production. The nutrient content and the hygienic quality of silage may affect both feed intake, weight gain and the number of foetuses in ewes. Fermentation quality in silage affects the feed intake in cows, but few experiments are done on sheep. Application of acidic additives increases aerobic stability of stability (Randby, 2002). After opening for feeding, air will usually penetrate via the silage face. Acid-tolerant aerobic microorganisms start to proliferate. This aerobic deterioration process is usually initiated by yeasts, which use residual sugars and lactic acid as substrates. When silage pH rises, *Bacillus* species and moulds proliferate, and growth of *Clostridia* may occur in anaerobic niches in the silage (Vissers *et al.*, 2007). Using silage exposed to air with no noticeable heating and moulding has so far not been tested on feed intake and production of sheep. The objectives of the experiment were to investigate the effects of silage additive, aerobic exposure of silage before feeding, and concentrate level, on feed intake and growth of ewe lambs.

Materials and methods

The experiment was carried out with 24 weaned ewe lambs from September to December. Average body weight (BW) at the start of the experiment was 36.2 (±4.8, 29.6-44.4) kg. The experiment was conducted as a 4×4 Latin square-design, with 4 silages, 6 squares and four 4-weeks periods. The four silage treatments were factorially arranged (2×2): (1) silage ensiled with or without acidic additive; (2) bale silage opened 0-2.5 days ('fresh') or 3.5-6 days ('old') before feeding. Two levels of concentrate (100 or 300 g day^{-1}) were offered in continuous design with 3 squares at each level.

The grass silage, dominated by timothy, was harvested on 29 May 2012. The grass was wilted over night before baling using an Orkel GP 1260 roundbaler with 20 fixed knives. Every second bale was added 5.5 l/ton GrasAAT Plus (58% formic acid, 12% propionic acid, 1.5% benzoic acid). The bales were wrapped in 8 layers of white plastic film. The concentrate used was FORMEL Sau (Felleskjøpet Agri, Norway). Grass silage was offered for *ad libitum* intake twice daily, together with concentrate. Individual feed intake was recorded seven days a week, and the ewe lambs were weighed for two consecutive days in week 1 and week 4 of each period. Bales were stored outdoors at declining mean temperatures from 10 to -5 °C, but moved indoors at 12 °C about a week before opening. Samples of 'fresh' bales were taken when plastic was removed and feeding initiated, and taken again of the same bales 3.5 and 6 days after opening, when considered 'old'. Heated or moulded silage was not offered to animals, and not included in feed samples.

Results and discussion

The analyses of silage showed that only one sample, 'old', without additive, had content above the reference value for good hygienic quality (*Clostridia* spores 2.3 log cfu g^{-1} (reference value 1.2 log cfu g^{-1}; Eurofins, 2015)). From bales compositing this sample some mould had been discarded. From 13% of the 'old' with additive, and 31% of the 'old' without additive, and only 3% of the 'fresh' bales, heated or moulded spots were detected and discarded. This indicated that after removing the heated and moulded parts, that constituted only a small portion of each heated or moulded bale, the remaining silage was mainly of good hygienic quality.

Additive treated silage had lower content of the fermentation products lactic acid, acetic acid, ethanol and NH_3-N, and higher content of water-soluble carbohydrates (WSC), than silage without additive (Table 1). Adding acidic additive to silage resulted in a restrictedly fermented silage, and no additions in a high lactate silage, as defined by Huhtanen *et al.* (2002). 'Fresh' silage had higher content of ethanol than 'old' silage. Otherwise, storage time of silage did not affect the chemical content.

The intake of DM, energy and protein was higher for lambs fed silage with than without additive (Table 2). Ewe lambs fed the highest level of concentrate had higher total DM intake. Intake of concentrate did not affect daily silage intake. Storage time of silage did not affect feed intake. The calculated relative silage DM intake index (Huhtanen *et al.,* 2007) was 113 and 106 for silage with and without additive, respectively (both 'fresh' and 'old' silage). A standard silage has an index of 100. The feed intake of the ewe lambs was 7% higher of silage with than without additive (0.98 and 0.91 kg DM), as expected according to the calculated intake index. The daily weight gain was higher for ewe lambs fed silage with than without additive (Table 2). There was no significant interaction between silage additive and time

Table 1. Composition of silage and concentrate (g kg^{-1} DM if nothing else given).[1]

	With additive		Without additive			*P*-value			Concentrate
	'Fresh'	'Old'	'Fresh'	'Old'	SEM	Additive	Storage	A×S	
DM g kg^{-1}	279	283	270	278	0.33	0.06	0.12	0.53	92.0
OM	921	921	921	920	0.85	0.50	0.65	0.54	930
Crude protein	141	143	145	148	1.73	0.05	0.20	0.85	167
NDF	502	500	501	503	6.15	0.82	0.99	0.74	228
WSC	66.7	65.3	11.7	10.7	4.25	<0.001	0.78	0.96	58.6
Lactic acid	45.8	46.8	91.3	94.7	3.40	<0.001	0.54	0.73	
Formic acid	11.0	10.7	0.91	0.44	0.87	<0.001	0.65	0.95	
Acetic acid	4.86	4.67	11.8	10.8	0.40	<0.001	0.18	0.36	
Propionic acid	2.18	2.09	0.03	0.00	0.17	<0.001	0.74	0.86	
Ethanol	9.95	7.40	16.5	11.6	0.41	<0.001	<0.001	0.02	
NH_3-N g kg^{-1} TN	56.2	54.2	73.1	77.4	1.72	<0.001	0.53	0.10	
pH	4.46	4.50	4.38	4.40	0.016	<0.001	0.11	0.60	
NE_L, MJ	6.62	6.62	6.66	6.65	0.032	0.34	0.94	1.00	7.48
AAT, g	71.2	71.1	71.2	71.2	0.13	0.63	0.77	0.92	130.1
PBV, g	18.1	20.1	21.5	24.3	1.74	0.06	0.20	0.84	21.7

[1] DM = dry matter; OM = organic matter; NDF = neutral detergent fibre; NE_L = net energy lactation; AAT = amino acids absorbed from the intestines; PBV = protein biological value; WSC = water-soluble carbohydrates; SEM = standard error of the mean.

Table 2. Feed intake and daily weight gain of ewe lambs.[1]

	Silage					Concentrate			*P*-value			
	With additive		Without		SEM	100 g	300 g	SEM	Additive	Storage	A×S	Conc.
	'Fresh'	'Old'	'Fresh'	'Old'								
Feed intake, day^{-1}												
Silage, kg DM	0.97^{a}	0.99^{a}	0.91^{b}	0.90^{b}	0.025	0.95	0.93	0.027	<0.001	0.76	0.40	0.44
Total, kg DM	1.15^{a}	1.17^{a}	1.10^{b}	1.09^{b}	0.025	1.05	1.21	0.027	<0.001	0.74	0.40	<0.001
AAT, g	93^{a}	94^{a}	89^{b}	88^{b}	1.79	80	102	1.9	<0.001	0.77	0.40	<0.001
NE_L, MJ	7.80^{a}	7.91^{a}	7.44^{b}	7.39^{b}	0.167	7.03	8.24	0.177	<0.001	0.76	0.40	<0.001
Weight gain, g day^{-1}	151^{a}	147^{a}	124^{ab}	97^{b}	10.2	115	144	8.4	<0.001	0.11	0.23	0.01

[1] DM = dry matter; NE_L = net energy lactation; AAT = amino acids absorbed from the intestines; SEM = standard error of the mean.

of storage (*P*=0.23). However, when acid was applied to the silage, the ewe lambs' weight gain increased numerically by 27 and 50 g day^{-1} for 'fresh' and 'old' silage, respectively. Increased concentrate level increased daily weight gain by 29 g day^{-1}.

Conclusions

Adding acidic additive to silage for weaned ewe lambs increased feed intake and daily weight gain. There was no main effect of aerobic storage time of silage before feeding. However, the use of acidic additive to silage tended to improve weight gain more when fed 'old' than 'fresh'. Increasing the level of concentrate from 100 to 300 g day^{-1} increased feed intake and weight gain. In general, heated and moulded silage should, regardless of the cause, never be fed to animals.

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The effect of delayed fertilizer N application on root biomass and N uptake of *Lolium perenne*

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Abstract

During the first days after harvest of *Lolium perenne* L., low N uptake from the soil may lead to N loss if N fertilizer is applied too soon. Furthermore, temporary N deprivation has been found to stimulate root growth. We therefore hypothesized that a strategic delay in N application after harvest may improve N use efficiency of *L. perenne* grassland by increasing root biomass and reducing N loss. In a laboratory and field experiment, we delayed N fertilizer application for 0, 3, 6, 9 and 12 days after harvest, and determined effects on herbage yield, herbage N uptake and root biomass of *L. perenne*. In both experiments, delaying N application with up to 12 days had no significant effect on root biFomass or total herbage N uptake. In the field experiment, total yield tended to be highest with a 3-day delay. For two harvests in the field experiment there was a significantly higher N uptake when N application was delayed, possibly due to rainfall-induced N losses in the treatments with shorter delay. Therefore, timing of N fertilizer application based on rainfall forecasts can contribute to improve N use efficiency by reducing N losses.

Keywords: N fertilization, delayed N application, root biomass, N use efficiency

Introduction

Strategic timing of N fertilizer application after cutting could be a valuable management tool to reduce N loss and increase N use efficiency of grassland, by synchronizing N supply with N demand. Various experiments have shown that during the first days of regrowth, most of the N comes from remobilized organic N stored in the stubble and roots, and that current root uptake of fertilizer N is low (Ourry *et al.*, 1989). Low uptake of fertilizer N during the first week after application increases the risk of N loss to the environment, especially with heavy rainfall. Additionally, there is evidence that temporary lack of N after harvest may stimulate root growth (Ennik and Hofman 1983; Jarvis and Macduff 1989). A higher rooting density and depth play a critical role in the interception and uptake of N by grass. The objective of the present study was to assess the effect of delaying N application after harvest on root biomass, herbage N uptake and herbage yield of *Lolium perenne* in a laboratory and field experiment. We hypothesized that temporary N deprivation by delaying N application after a harvest event may increase root biomass of *L perenne* resulting in increased N uptake and possibly also herbage yield.

Materials and methods

In the laboratory experiment, undisturbed *L. perenne* grassland soil cores (40 cm depth, 11.5 cm diameter) were placed in a controlled growth room. The grass was cut every 21 days, resulting in four harvests. N fertilizer application (100 kg N ha^{-1} cut^{-1}) was delayed for either 0, 3, 6, 9 or 12 days after each cut. Every four days a 10 mm rainfall event was simulated.

In the field experiment, we tested the same N delay treatments as in the laboratory experiment. The field experiment had a randomized block design with six replicate plots (10×2.5 m) in six locations (blocks) of permanent *L. perenne* grassland on a drought-sensitive sandy soil. The experiment included six harvest cycles (growth periods), with 4 to 6 weeks between each harvest. Total N application was 320 kg N ha^{-1} yr^{-1}. No irrigation was given.

In both experiments, herbage yield and N uptake were determined by cutting the grass to 6 cm height and measuring DM weight and N content. Roots were sampled after the final harvest. In the laboratory experiment, all roots per core per layer of 10 cm depth (0-10, 10-20, 20-30 and 30-40 cm) were sampled. In the field experiment, roots were sampled by taking three soil cores (8.5 cm diameter) from three soil layers (10-20, 20-30 and 30-40 cm depth, excluding 0-10 cm depth) in each plot. All soil cores were carefully washed through a 2 mm mesh screen and oven-dried at 70 °C for 24 hours to determine root biomass.

Results and discussion

In contrast to our hypothesis, delayed N application had no significant effect on total root biomass. A significant treatment effect on root biomass was only found for the 20-30 cm soil layer in the field experiment ($P<0.05$, Figure 1). In the experiments by Ennik and Hofman (1983) and Jarvis and Macduff (1989), treatments consisted of controlled interruptions of a continuous N supply to grass grown in nutrient solution, whereas in our experiments, N was supplied only at the start of each growth period. In our experiments it is possible that applied N was depleted before the next harvest and that in some cases all delay treatments may have experienced N shortage, masking effects of delayed N application on root biomass.

In the laboratory experiment, total herbage yield (cumulative over all harvests) was highest after a 3-day delay and was significantly reduced when N application was delayed by 6 days or more, with lowest yields observed after a 12-day delay (Table 1). A similar, though non-significant trend, was observed for total herbage yield in the field experiment (Table 1). This difference in significance may be explained by the shorter harvest cycles in the laboratory experiment (three weeks) compared to the field experiment (four to six weeks), which potentially increased the impact of delayed N application on herbage yield by reducing the opportunity for conversion of N into biomass in the former.

In the laboratory experiment, total herbage N uptake ranged from 247 to 304 kg N ha^{-1} (Table 1), and was not significantly affected by delaying N application. Similarly, in the field experiment, no significant effect of delay treatments on total herbage N uptake was detected (Table 1). However, when analysing the data for individual growth periods, we found that N uptake during the fourth and fifth period in the

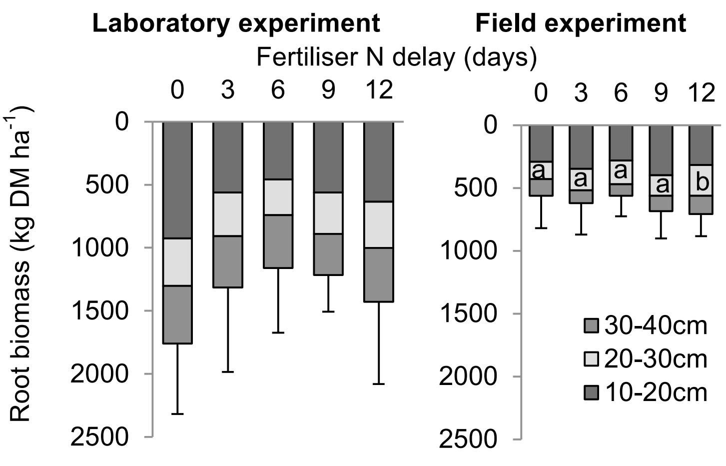

Figure 1. Mean *Lolium perenne* root biomass (kg DM ha^{-1}) in soil layers 10-20, 20-30 and 30-40 cm, as affected by delaying N application after harvest with 0, 3, 6, 9, or 12 days. Error bars represent standard errors of total root biomass (soil layer 10-40 cm). Different letters in treatment means within a soil layer indicate a significant difference (least significant difference 5%).

Table 1. The effect of delayed N application (0, 3, 6, 9 or 12 days) on herbage dry matter yield (dry matter yield (DMY), ton dry matter ha^{-1}) and N uptake (kg N ha^{-1}) in the laboratory and field experiment.[1]

	Laboratory experiment		Field experiment					
Delay	**DMY**	**N uptake**	**DMY**			**N uptake**		
(days)	Total[2]	Total	H4	H5	Total[2]	H4	H5	Total
0	18.7^{cd}	247	1.7	1.4^{a}	11.1	46^{a}	39^{a}	224
3	19.8^{d}	290	1.8	1.6^{b}	11.4	46^{a}	49^{b}	239
6	15.7^{bc}	277	1.8	1.5^{b}	11.1	48^{a}	47^{b}	238
9	15.1^{ab}	298	1.8	1.5^{ab}	10.7	51^{ab}	48^{b}	236
12	12.1^{a}	304	2.0	1.5^{b}	10.5	59^{b}	50^{b}	241
LSD[3] (P<0.05)	3.5	52	275	0.13	8.25	8	6	25
P-value	<0.01	0.19	0.11	0.04	0.18	0.01	0.01	0.65

[1] Different letters within a column indicate a significant (P<0.05) difference between treatments.

[2] Cumulative across 4 harvests for the laboratory experiment and 6 harvests for the field experiment; H4 = harvest 4; H5 = harvest 5.

[3] LSD = least significant difference.

field experiment was significantly higher (P=0.01) in plots where N application had been delayed with 12 days (fourth growth period) or 3 days or more (fifth growth period) (Table 1). This was probably related to rainfall patterns during the first 12 days of these growth periods. The significantly higher N uptake after N application delay of 9 to 12 days in the fourth period of the field experiment was associated with avoidance of N losses through leaching, high levels of rainfall occurred on day 6, 8 and 10 of that period. Similarly, the positive delay effect in the fifth period was associated with heavy rainfall on day 1 during that period. The apparent relatively large effects of rainfall events on herbage N uptake and herbage yield observed in our experiment suggest that delay of fertilizer N application can be used as a tool to increase N use efficiency and herbage yield of grassland by minimizing the risk of fertilizer N leaching.

Conclusions

In our experiments, a delay in N application of up to 12 days after harvest had no significant effect on *L. perenne* root biomass or total N uptake, but did significantly reduce total herbage yield in the laboratory experiment. The observed positive effects of delayed N application on N uptake and herbage yield in the fourth and fifth growth period in the field experiment appear to be the result of the avoidance of leaching, caused by heavy rainfall shortly after fertilizer N application. This suggests that strategic timing of N application based on rainfall forecasts could contribute to reduce N losses from leaching.

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Including bioactive legumes in grass silage to improve productivity and reduce pollutant emissions

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Abstract

The aim of this study was to quantify the benefits of including in grass-based silages two bioactive legume species, namely *Onobrychis viciifolia* (sainfoin, SF) and *Trifolium pratense* (red clover, RC) containing condensed tannins (CTs) or polyphenol oxidase, respectively. We conducted an experiment in small scale silos and two *in vivo* experiments using silages of different mixtures with a grass species (*Phleum pratense* L., timothy, T, control) to investigate silage quality and conservation, lambs' performance, digestive efficiency, nitrogen (N) balance and methane (CH_4) emissions in sheep. Ensiling bioactive legumes in mixtures with grass improved fermentation and proteins' protection from degradation within the silos. Due to a different fibre composition and the presence of CTs which impaired SF digestibility, lambs that consumed T-SF mixture showed lower intake and performance than those that received RC-containing silages. An environment-friendly shift in N excretion from urine to faeces and a reduction in CH emissions when animals were fed with SF-containing mixtures were also observed.

Keywords: *Trifolium pratense*, *Onobrychis viciifolia*, silage quality, animal performance, protein protection, methane emissions

Introduction

Fodder legume species can reduce inputs in livestock production systems (fertilizer and rich-protein concentrates) notably because they contain high levels of crude proteins, which are of primary importance in ruminant nutrition. However, during both silage and rumen fermentation processes, proteins are submitted to degradation which affects forage nutritive value and leads to nitrogen (N) losses, notably via urine. Some particular legumes may have a particular interest as they produce bioactive compounds that can positively affect silage quality and digestive processes. Condensed tannins (CTs) present in *Onobrychis viciifolia* (sainfoin, SF) are able to bind proteins leading to diverse effects on ruminant nutrition and health (Mueller-Harvey, 2006). *Trifolium pratense* (red clover, RC) contains polyphenol oxidase (PPO), an enzyme that catalyses the oxidation of different phenolics into quinones. Similar to CTs, quinones are able to form complexes with proteins that could similarly reduce their degradation in the silo and the rumen (Lee, 2014). The aim of this work was to investigate and quantify the potential benefits of using these two bioactive legume species on (1) quality and conservation of silages, (2) digestive efficiency and sheep performance, and (3) environmental footprint (N excretion and CH emissions).

Materials and methods

Plants (T, SF and RC) were grown separately at INRA site of Crouël (altitude 320 m, central France). On 9-11 May 2012, plants (35.6 g CTs kg^{-1} dry matter (DM) in SF; PPO activity in RC: $\Delta OD\ min^{-1} = 4.9$) were harvested during the first vegetation cycle for an experiment in small scale silos to assess silage quality and conservation. Plants were chopped, wilted and ensiled in triplicate at 25% DM as pure forages, binary (50% T + 50% SF, 50% T + 50% RC) and ternary mixtures (50% T + 25% SF + 25% RC) (see details in Copani *et al.*, 2014). On 2 October 2012, silos were opened and sampled to determine pH, and DM, neutral detergent fibre (NDF), crude protein (CP), lactic acid, soluble N and NH concentrations in silage. The next year, two *in vivo* experiments were simultaneously conducted to measure (1) lambs' performance through measurements of voluntary intake, weight gain and feed conversion efficiency (FCE) (see details including silage characteristics in Copani *et al.*, 2016), and (2) digestion parameters, N balance and CH emissions (SF technique) using 10 young sheep fed according to a repeated Latin square design. The treatments tested *in vivo* were pure T, 50-50% binary mixtures (T-SF, T-RC, SF-RC) and the ternary mixture (50% T + 25% SF + 25% RC), fed with a restricted barley supplement for growing lambs.

Results and discussion

The silage characteristics of silages in small scale silos (Table 1) showed clearly that using bioactive legumes as pure forage or in mixtures with grass improved the fermentation process as indicated by the lower pH (P=0.011) and greater lactic acid concentration (P=0.001) compared to pure grass silage, but

Table 1. Chemical composition and quality of conservation of small scale silos.

Item[1]	Pure plants[2,3]			Mixtures[2,3]			SEM	P-value
	T	SF	RC	T-SF	T-SF-RC	T-RC		
DM, g kg^{-1}	247	243	277	251	271	265	15.3	0.456
NDF, g kg^{-1} DM	570^{a}	334^{c}	291^{c}	452^{b}	450^{b}	423^{b}	8.8	<0.001
CP, g kg^{-1} DM	173^{e}	222^{ab}	234^{a}	194^{d}	201^{cd}	209^{bc}	6.0	<0.001
pH	5.2^{a}	4.3^{b}	4.5^{b}	4.4^{b}	4.0^{b}	4.5^{ab}	0.14	0.011
Lactic acid, g kg^{-1} DM	44.5^{c}	89.7^{ab}	102.2^{a}	68.9^{bc}	94.1^{ab}	81.4^{ab}	4.73	0.001
Soluble N, % total N	54.6^{a}	42.0^{bc}	32.6^{c}	43.3^{b}	36.1^{bc}	37.1^{bc}	2.14	<0.001
NH, % total N	16.8^{a}	13.6^{ab}	7.6^{b}	9.1^{ab}	8.1^{b}	8.1^{b}	1.56	0.009

[1] DM = dry matter; NDF = neutral detergent fibre; CP = crude protein.

[2] T = *Phleum pratense* L., timothy; SF = *Onobrychis viciifolia*, sainfoin; RC = *Trifolium pratense*, red clover.

[3] Within a row, means followed by different letters are significantly different (P<0.05). SEM = standard error of the mean.

Table 2. Animal performance data of lambs and digestion parameters of young sheep fed with the experimental silages.

Item[1]	Silages[2,3]					SEM	P-value
	T	T-SF	T-SF-RC	T-RC	RC-SF		
Performance experiment							
DM intake, kg d^{-1}	1.056^{b}	1.021^{b}	1.186^{a}	1.209^{a}	1.247^{a}	0.0378	<0.001
ADG, kg LWG d^{-1}	0.181^{bc}	0.145^{c}	0.222^{a}	0.235^{a}	0.214^{ab}	0.0129	<0.001
FCE, kg LWG kg^{-1} DM intake	0.175^{a}	0.147^{b}	0.192^{a}	0.198^{a}	0.177^{a}	0.0098	0.009
Final LW (week 10), kg	40.36^{b}	38.64^{b}	43.89^{a}	43.87^{a}	43.92^{a}	0.831	<0.001
Cold carcass weight, kg	18.63^{b}	17.23^{c}	20.13^{a}	20.48^{a}	20.49^{a}	0.518	<0.001
Digestion experiment							
OM digestibility, %	74.4^{a}	69.9^{bc}	70.9^{bc}	73.5^{ab}	70.0^{c}	1.06	0.002
NDF digestibility, %	76.0^{a}	57.4^{c}	64.5^{b}	71.5^{a}	53.9^{c}	1.50	<0.001
N digestibility, %	67.9^{a}	61.2^{b}	65.5^{ab}	68.8^{a}	65.1^{ab}	1.83	0.003
N faeces, g g^{-1} N intake	0.321^{b}	0.388^{a}	0.345^{ab}	0.312^{b}	0.349^{ab}	0.0183	0.003
N urine, g g^{-1} N intake	0.461	0.396	0.407	0.450	0.422	0.0247	0.108
N retained, g d^{-1}	5.96	6.91	9.17	8.20	9.17	1.178	0.067
CH emissions, g kg^{-1} DM intake	35.7^{a}	29.7^{b}	29.3^{b}	30.5^{ab}	27.2^{b}	1.06	0.003

[1] DM = dry matter; OM = organic matter; NDF = neutral detergent fibre; ADG = average daily gain; LWG = live weight gain; FCE = feed conversion efficiency.

[2] T = *Phleum pratense* L., timothy; SF = *Onobrychis viciifolia*, sainfoin; RC = *Trifolium pratense*, red clover.

[3] Within a row, means followed by different letters are significantly different (P<0.05). SEM = standard error of the mean.

also protected the integrity of protein as indicated by lower soluble N and ammonia concentrations (P<0.001 and P=0.009, respectively).

The *in vivo* experiments showed that the presence of RC within silage can lead to greater DM intake (P<0.001) and weight gain (P<0.001) than for T or T-SF (Table 2). This could be due to differences in fibre profiles between SF and RC and the presence of CTs which led to lower OM and NDF digestibility of SF- than RC-containing silages (P=0.002 and P<0.001, respectively). When animals were fed with SF-containing mixtures, a shift in N excretion from urine to faeces (from which N is less volatile than in urine) and a reduction in CH emissions compared to pure grass (P=0.003) were also observed.

Conclusions

Under our experimental conditions, RC appeared to provide greater benefit for animal production, while SF allowed lowered N and CH wastes.

Acknowledgments

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Meta-analysis of the impact of white clover inclusion on milk production of grazing dairy cows

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Abstract

There is recent interest in the introduction of white clover (WC) in Ireland to improve the efficiency of pasture-based grazing systems. After a literature review, a meta-analysis was undertaken to investigate the effect of WC inclusion in perennial ryegrass swards on milk production. For analytical purposes two databases were created, containing 15 and 20 papers, respectively, published between 1985 and 2015. Database one, contained experiments comparing milk production from cows that grazed on grass-only (GO) and grass-clover (GC) swards. Database two contained only experiments that had GC swards to investigate the impact of varying sward WC content on milk production. In database one, the mean WC content of the GC swards was 33.6%. Mean daily milk yield was greater (P<0.001) for cows that grazed on GC swards compared with GO swards (18.8 kg vs 17.6 kg, respectively). However, stocking rate and milk yield per hectare were reduced (P<0.05) for the GC swards. In conclusion, the inclusion of WC into perennial ryegrass swards increased milk yield per cow but reduced milk yield per hectare.

Keywords: meta-analysis, white clover, dairy cow, milk production, grazing

Introduction

There is increased demand for dairy products worldwide (Delgado, 2005), which is coupled with the realisation that consumers want dairy products that are produced in a sustainable and environmentally benign manner. Traditionally white clover (*Trifolium repens* L., WC) was included in perennial ryegrass (*Lolium perenne* L., PRG) swards as a means of improving sward nutritive value and reducing nitrogen (N) use. However, intensification in the use of cheap mineral N has resulted in a reduction in the use of WC. Forage legumes, and WC in particular, can make an important contribution to the sustainability of pasture-based ruminant production systems (Peyraud *et al.,* 2009). Therefore, the interest in using forage legumes is increasing again because they offer opportunities for sustainable pasture-based production systems by (1) increasing pasture yield, (2) substituting inorganic N-fertiliser inputs with symbiotic N2 fixation, (3) mitigating and facilitating adaptation to climate change and (4) increasing the nutritive value of pasture and raising the efficiency of conversion of pasture to animal protein (Lüscher *et al.,* 2014). Therefore, the objective of this study was to quantify the milk production response associated with the introduction of WC into a PRG sward.

Materials and methods

An electronic literature search was undertaken to identify papers where the effect of WC inclusion on milk production in lactating dairy cows was studied. Two separate databases were created. In database one, papers were selected if they contained a comparison of milk production from lactating dairy cows grazing PRG-WC (GC) swards against PRG only (GO) swards. Experimental characteristics required were experiment length, stocking rate, grazing cow-days ha^{-1}, WC content of the sward and milk production results per cow and/or per ha. In experiments where a sub-factor was studied (e.g. 2 supplementation levels or 2 post-grazing heights) or multiple years of data were reported, comparisons of milk production from GC and GO swards conducted under similar experimental conditions were considered independent studies. If a treatment had sward WC content below 5% in an experiment, then it was considered as a GO treatment. Each comparison of milk production from GC and GO swards was allocated by an individual experimental code (study). Database one contained 15 papers and 43 comparisons of milk production from GC and GO swards published between 1985 and 2015. Data from database one was analysed using mixed models (PROC MIXED) in SAS version 9.3 (SAS institute, Cary, NC, USA). Terms included in the model were treatment (GC and GO) and study effect, which represented the variance between studies not accounted for by the variables in the model, as described by Sauvant *et al.* (2008). In database two, papers were selected if they contained milk production from lactating dairy cows grazing on GC swards with varying levels of WC content. Database two was constructed similarly to database one. Database two contained 20 papers and 87 data points of milk production from GC swards published between 1989 and 2015. Data from database two was also analysed using mixed models (PROC MIXED) in SAS. Terms included in the model were WC content and the study effect. Database two experiments reflect the variation in milk production as WC content increases in the sward.

Results and discussion

The mean experimental characteristics and milk production per cow and per ha for database one were; number of cows: 49, stocking rate: 3.12 cows ha^{-1}, experiment length: 119 days, N application: 139 kg ha^{-1}, WC content: 17.1%, milk yield cow^{-1}: 18.2 kg and milk yield ha^{-1}: 7,876 kg. The average effect of introducing WC into a sward on experimental characteristics and milk production per cow and per ha for database one is presented in Table 1. Mean daily milk and milk solids (fat + protein) yield per cow were greater ($P<0.001$) by 1.2 and 0.1 kg cow^{-1} for cows that grazed on GC swards compared with GO swards. The presence of WC in the sward did not affect fat or protein content, however, lactose content was greater for cows that grazed on GC swards. Stocking rate was reduced by 0.29 cows ha^{-1} on the GC swards, and consequently, milk and milk solids yield per ha were reduced ($P<0.05$) by 431 and 35 kg ha^{-1}, respectively. The mean experimental characteristics and milk production per cow and per ha for database two were; number of cows: 49, stocking rate: 2.82 cows ha^{-1}, experiment length: 148 days, N application: 76 kg ha^{-1}, WC content: 24.8%, milk yield cow^{-1}: 19.2 kg and milk yield ha^{-1}: 10,544 kg. The equations that accounted for the greatest proportion of variation in predicted milk production per cow and per ha according to WC content were linear, with the exception of milk solids yield and protein content per cow and milk and milk solids yield per ha, which were quadratic (results not shown). The residual standard error was low for milk yield (0.95), which indicates a good precision of the predictive equations. On the basis of the predictive equations, a 10% increase in sward WC content resulted in a mean proportional increase of 1.46 and 0.97% for daily milk and milk solids yield, respectively, whereas milk and milk solids yield per ha were reduced by 15.3 and 12.0%, respectively. This is in accordance with Harris *et al.* (1997) who stated that as sward WC content increased, daily milk yield per cow also increased. Within grazing systems, stocking rate, milk yield per cow and milk yield per ha are closely linked (McCarthy *et al.,* 2011). Generally, as stocking rate increases milk yield per cow decreases and milk yield per ha increases (Macdonald *et al.,* 2008) and vice versa. The increase in daily milk yield per cow for cows grazing on GC swards was partly due to the reduction in stocking rate and partly due to the presence of WC in the sward. Using the stocking rate effect prediction equations of McCarthy *et al.* (2011), it was calculated that the increase in milk yield per cow was 38% due to the reduction in stocking rate and 62% due to the presence of WC in the sward. However, despite the increase in daily milk yield per cow, milk yield per ha decreased with GC swards. Grazing experiments that have compared GO and GC swards, have reduced stocking rate and N application rates on the GC swards and as a consequence GC systems had lower levels of productivity. In the context of increased demand for dairy products, this raises the question as to the possibilities of incorporating WC into more intensive pasture-based production systems.

Table 1. Effect of introducing white clover into a perennial ryegrass sward on milk production per cow and per ha for database one data.

	No. data	Grass-only	Grass-Clover	SE[1]	*P*-value
Experimental characteristics					
Clover (%)	86	0.6	33.6	2.38	<0.001
Stocking rate (cows ha^{-1})	66	3.27	2.98	0.270	<0.001
Nitrogen (kg ha^{-1})	66	206	72	21.6	<0.0001
Grazing days ha^{-1}	54	457	418	36.6	<0.001
Production per cow					
Milk yield (kg cow^{-1})	86	17.6	18.8	0.70	<0.001
Fat content (g kg^{-1})	74	42.0	41.5	1.01	0.171
Protein content (g kg^{-1})	74	33.6	33.9	0.61	0.194
Lactose content (g kg^{-1})	32	44.6	45.3	0.79	0.004
Milk solids[2] yield (kg cow^{-1})	82	1.31	1.41	0.046	<0.0001
Production per hectare					
Milk yield (kg ha^{-1})	54	8092	7661	775.5	0.017
Milk solids yield (kg ha^{-1})	50	618	583	64.9	0.012

[1] SE = standard error.

[2] Milk solids = kg of fat + protein.

Conclusions

Mean daily yield of milk and milk solids increased when WC was included in a PRG sward. However, due to management associated with WC swards (i.e. reduced stocking rate and N application rates) milk and milk solids yield per ha were reduced. The results raise interesting questions as to the possibilities of incorporating WC into more intensive pasture-based production systems and further research is required in this area.

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The effect of grazing multispecies swards on lamb performance and herbage production

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Abstract

Multispecies swards have potential for increased biomass production at lower nitrogen (N) inputs and thus have potential to reduce N requirements in pasture-based lamb production systems. Four sward treatments were investigated in a farmlet study; perennial ryegrass (*Lolium perenne*) only (PRG), receiving 163 kg N per hectare per year (kg N/ha/y); PRG and white clover (*Trifolium repens*) mix at 90 kg N ha^{-1} y^{-1} (PRG+WC); six species mix at 90 kg N ha^{-1} y^{-1} (6S) and a nine species mix at 90 kg N ha^{-1} y^{-1} (9S). Farmlets were stocked with 30 twin-rearing ewes at a stocking rate of 12.5 ewes ha^{-1} (2.5 livestock units (LU) ha^{-1} pre-weaning and 5.5 LU ha^{-1} post weaning) from four weeks post-partum until lambs were slaughtered at 45 kg liveweight under rotational grazing management. Lambs were weighed fortnightly and average daily gain (ADG) calculated. Pasture herbage mass was measured before and after each grazing event by taking three quadrat (0.5×0.5 m) cuts and harvesting to 4 cm. The harvested herbage was weighed and retained for DM determination. Data was analysed using Proc Mixed SAS 9.4. Compared to lambs grazing PRG, lambs grazing the 6S and the 9S had higher ADG from birth to slaughter (279, 260 compared to 238 g day^{-1}) respectively, P<0.01), higher weaning weights (36.3 and 34.4 compared to 32.3 kg, respectively, P<0.01) and subsequently reduced days to slaughter (151, 161 and 176 days, respectively, P<0.01). To conclude, the 6S and 9S mixes supported higher animal performance than PRG swards and there was no significant effect of sward treatment on annual herbage production.

Keywords: multispecies swards, perennial ryegrass, lamb performance, lamb average daily gain

Introduction

Grass-based livestock systems largely rely on the production of high quality forage to support animal performance. As grass has the lowest production cost of all feeds (Finneran *et al.*, 2010) available, it is the key component of profitable pasture based sheep production systems. Daily liveweight gain targets of 295 g day^{-1} for twin suckling lambs pre-weaning from grazed grass only are set in Irish sheep production (Diskin and McHugh, 2012). Multispecies swards produce higher quantities of dry matter (DM) from lower N inputs (Nyfeler *et al.*, 2009), while having a higher feeding value for grazing animals (Kemp *et al.*, 2010) thus representing a potential alternative to PRG only swards. The objective of this study was to examine the effect of grazing multispecies swards compared to PRG only swards on lamb performance and herbage production under intensive rotational grazing management.

Materials and methods

A complete randomised block design was used to investigate the effect of four sward treatments on lamb performance and herbage production. Four experimental farmlets were established at UCD Lyons Research Farm (53°17 N, 6°31 W, and 80.5 m above sea level) in early September 2014. The four treatments were as follows: (1) PRG receiving 163 kg N ha^{-1} y^{-1} (PRG); (2) PRG and white clover receiving 90 kg N/ha/y (PRG+WC); (3) a six species mix containing two grasses (PRG and timothy (*Phleum pratense*), two legumes (white clover and red clover (*Trifolium pratense*) and two herbs (ribwort plantain (*Plantago lanceolata*) and chicory (*Cichorium intybus*) receiving 90 kg N ha^{-1} y^{-1} (6S); (4) a nine species mix containing three grasses (PRG, timothy and cocksfoot (*Dactylis glomerata*) three legumes (white clover, red clover and greater birdsfoot trefoil (*Lotus penduncculatus*)) and three herbs (ribwort plantain, chicory and yarrow (*Achillea millefolium*)) receiving 90 kg N ha^{-1} y^{-1} (9S). Each farmlet consisted of 2.4 ha (5 paddocks) which were rotationally grazed at stocking rate of 12.5 ewes ha^{-1} (2.5 livestock units (LU) ha^{-1} pre-weaning and 5.5 LU post weaning) or 30 twin-rearing ewes per treatment. The target pre-grazing herbage mass was 1,200 kg DM ha^{-1} (cut 4 cm above ground level) for the duration of the experiment and the target for post grazing sward height (PGSH) was 4 cm. Herbage mass was recorded before and after each grazing event by taking three quadrat (0.5×0.5 m quadrat) cuts and harvested to 4 cm. The harvested herbage was weighed and retained for DM determination. Lambs were weaned at 14 weeks old. Post weaning, a leader follower system was operated with lambs grazing ahead of the ewes. Lambs were removed from paddocks upon achieving a PGSH of 5 cm, with ewes introduced to achieve a PGSH of 4 cm. Herbage samples were taken prior to each grazing event and were separated to functional group (grass, legume and herb) by manual separation. Lambs were weighed fortnightly using a portable electronic scales (Prattley, Temuka, New Zealand), liveweight recorded electronically (Tru-Test Group, Auckland, New Zealand) and were drafted for slaughter on reaching 45 kg of liveweight. Data was analysed as a completely randomized block design using mixed models, where the ewe was the experimental unit (30 experimental units per treatment), with block of the random effect (PROC MIXED) in SAS (SAS, version 9.4, Inst. Inc., Cary, NC, USA).

Results and discussion

The effect of treatment on weaning weight, the ADG from birth to weaning and birth to slaughter, days to slaughter, kill out percentage and carcass weight are presented in Table 1. Lambs grazing the 6S sward treatment had a higher weaning weight (36.3 kg) compared to all other treatments (P<0.01), while lambs offered the PRG sward treatment had lower weaning weights than the 9S (P<0.01) sward treatment. Lambs grazing the 6S sward treatment had higher growth rates from birth to weaning compared to PRG and 9S (*P<0.05*) with PRG+WC intermediate. Both the 6S and 9S mixtures supported higher ADG from birth to slaughter in comparison to PRG (P<0.01), with the 6S treatment also recording higher birth to slaughter ADG compared to PRG+WC (P<0.01). Kill out percentage and subsequently carcass weight were unaffected by treatment.

Table 1. The effect of sward treatment on lamb weaning weight (kg), lamb growth rates from birth to weaning and birth to slaughter, days to slaughter, kill out percentage, carcass weight (kg), annual herbage production (kg DM ha^{-1}) and the number of times each sward treatment was grazed (LSM±SEM). The mean annual proportions (% of sward) of grass, legume and herb in the four sward treatments.[1]

	PRG	PRG+WC	6S	9S	SEM	*P*-value
Weaning weight (kg)	32.3^{a}	34.0^{ac}	36.3^{d}	34.4^{bc}	0.77	<0.01
Growth rate from birth to weaning (g day^{-1})	285^{a}	291^{ab}	303^{b}	286^{a}	8.3	<0.05
Growth rate from birth to slaughter (g day^{-1})	238^{a}	257^{ac}	279^{b}	260^{c}	8.3	<0.01
Growth rate from weaning to slaughter (d day^{-1})	201	205	223	202	12.5	NS
Days to slaughter	176^{a}	165^{ac}	151^{b}	161^{bc}	5.1	<0.01
Kill out %	44.5	44.5	44.2	44.3	0.45	NS
Carcass weight (kg)	20.3	20.7	20.2	20.3	0.22	NS
Annual herbage production (kg DM ha)	6107.0	6093.5	5847.9	5646.6	413.38	NS
Grass content (% of sward)	95.98^{a}	90.31^{b}	75.52^{cd}	79.06^{cd}	3.137	<0.05
Legume content (% of sward)	1.05^{a}	6.49^{bc}	5.59^{abc}	3.41^{a}	1.327	<0.05
Herb content (% of sward)	2.98^{a}	3.09^{a}	19.85^{bc}	17.53^{bc}	2.254	<0.05
Number of times grazed	6.2^{a}	6.4^{ab}	5.8^{a}	7.2^{ab}	0.35	<0.05

[1] DM = dry matter; PRG = perennial ryegrass; WC = white clover; 6S = six species mix; 9S = nine species mix; LSM = least significant mean; SEM = standard error of the mean.

The effect of treatment on annual herbage production, number of grazings per sward treatment and on annual proportional representations of the individual functional groups in each treatment are presented in Table 1. The PRG only sward which received 163 kg N ha^{-1} yr^{-1} produced the same quantity of herbage DM as the PRG+WC, the 6S and the 9S mixes which received 90 kg N ha^{-1} yr^{-1}. The PRG and 6S paddocks were grazed less times throughout the year than the PRG+WC and the 9S treatments ($P<0.05$).

Conclusions

Lambs grazing the 6S showed increased animal performance compared to PRG only swards in year one of a two year study. Lambs grazing the 6S swards had higher weaning weights due to higher pre-weaning ADG. There was no difference in the annual DM production from the mixtures (PRG+WC, 6S and 9S) despite receiving a lower N input compared to the PRG sward. The experiment will be repeated in 2016 before final conclusions can be drawn.

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Evaluation of perennial ryegrass variety performance on Irish farms

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Abstract

Increasing grass utilisation on-farm has favourable consequences for the environmental and financial sustainability of Irish farms. It is essential that a robust evaluation process exists to identify suitable grass varieties that perform well on commercial farms. Plant breeders aim to develop varieties which excel within recommended list evaluations but ultimately variety performance needs to be quantified in 'real life' commercial situations. Current plot-based evaluations expose cultivars to minimal stress under controlled test procedures. On-farm variety evaluations have the ability to determine the true agronomic potential of varieties due to greater exposure to stress imposed across a wider array of environments and management systems. The objective of this study was to establish the phenotypic dry matter (DM) yield performance of a range of perennial ryegrass varieties under commercial farm conditions. This study and future on-farm variety evaluations will provide improved direction to plant breeders in developing superior varieties with enhanced on-farm performance. For this study, monocultures of varieties were sown on 68 commercial farms across Ireland with a control variety established on each farm. Varieties were evaluated over 3 years. No significant difference in total DM yield existed among varieties although the range between the highest and lowest yielding varieties was 1.3 t DM ha^{-1}. On-farm evaluations allow for improved understanding of traits which can affect grass utilisation and lifetime productivity.

Keywords: *Lolium perenne*, on-farm, variety, evaluation

Introduction

Grazed grass is the lowest cost feed available to ruminant production systems in Ireland (Finneran *et al.*, 2010). Irish producers who operate predominantly pasture-based production systems enjoy a competitive advantage over those who operate higher intensity confinement based systems, as cost per unit output is reduced. There has been a surge in the level of interest in pasture-based systems in most temperate regions as a direct result of increased price volatility for both inputs and outputs (Dillon *et al.*, 2005). Central to the success of pastoral systems is the selection of the appropriate grass variety suited to localised environments.

The current recommended list evaluation process in Ireland involves all candidate varieties being evaluated for 2 full growing seasons. Following an establishment year they're assessed for dry matter (DM) production, nutritive value and persistency (Grogan and Gilliland, 2011). Within this process, variety suitability for animal grazing is evaluated by means of simulated grazing using mechanical harvesters. However, the stress levels imposed upon varieties is not representative of that at commercial farm level. Given the importance of grass to the sustainability of ruminant enterprises, it would be highly beneficial to have a robust and representative evaluation system, capable of identifying the elite performing varieties able to perform across a range of different farm practices. As highlighted by Stewart and Hayes (2011), recommended list evaluations strongly dictate the emphasis breeders place on traits. Thus if traits of economic relevance can be integrated into on-farm evaluations this should lead to the development of varieties with enhanced value for commercial performance.

On-farm variety evaluation is a new and innovative approach with the capability of identifying the true agronomic potential of ryegrass varieties. The development of a comprehensive national grassland database PastureBaseIreland (PBI) (http://www.pasturebase.teagasc.ie) has made on-farm evaluations possible, facilitating the collation of information on individual paddocks. The objective of the present study was to quantify the differences in grass DM yield of perennial ryegrass varieties on a large number of Irish farms as part of an on-farm variety evaluation process.

Materials and methods

The on-farm evaluation process began with the establishment of monocultures from a range of perennial ryegrass varieties. Varieties were sown in individual paddocks from 2011 to 2014 at a rate of 34.5 kg ha^{-1}. The varieties sown were: AberChoice (D), AberGain (T), AberMagic (D), Astonenergy (T), Drumbo (D), Dunluce (T), Glenveagh (D), Kintyre (T), Majestic (D), Twymax (T) and Tyrella (D); (D – diploid), (T – tetraploid). Tyrella was established on each farm as a standard control variety. Records from 68 predominantly dairy farms were used to evaluate the varieties. Farms on which varieties were sown are across a range of agroclimatic regions on varied soil types and management systems. Variety productivity was measured during the years 2013, 2014 and 2015. Over the 3 years a total of 900 individual paddock records were used to calculate the productivity of varieties. DM production was measured from 1 Jan-10 Dec each year. Grazing and silage yields were assessed prior to actual grazing or the conservation harvest. This estimate of herbage mass in each paddock was assessed visually and calibrated by cutting and weighing (O'Donovan *et al.*, 2002) or by measurement using a rising plate meter (McSweeney *et al.*, 2015). Only farms with greater than 30 complete farm grass measurements in each year were included in the dataset.

Least squares means for the different varieties were estimated using mixed models; paddock nested within farm was included as a repeated measure with a compound symmetry covariance structure assumed among paddocks within farm. The dependent variable was total paddock yield (kg DM ha^{-1}). Fixed effects considered in the mixed model were farm, sward age, year, variety and year by variety. Farm paddock was the experimental unit with farm acting as the replicate for the purposes of this study.

Results and discussion

Production differed significantly between farms. However this had a substantial effect on the statistical model as a repeated measure was required to isolate 'background noise' in order to reveal any varietal production differences. It was also found that sward age, year and year by variety had a significant effect on total DM production. Despite all these variable factors the range in DM yield between the highest and lowest yielding variety on-farm was 1.3 t DM ha^{-1}. However this overall mean difference was not significant nor were any of the varieties found to be significantly different in performance on-farm (P=0.425). Assessing variety persistence based on yields also proved difficult as yearly yield varied in descending order from 2015, 2014 and 2013 which was directly attributed to differences in meteorological conditions and maturing swards approaching the age of peak production (Figure 1).

The number of grazings achieved per variety, a trait which is unquantifiable under simulated grazing studies, was also recorded and an additional grazing was achieved between the varieties with the highest and lowest numerical yields. The absence of significant variety differences was not entirely due to increased on-farm variability as the higher performing varieties such as Tyrella had comparable levels of variability (standard error = 0.202 t DM ha^{-1}) to that achieved in plot based evaluations.

Conclusions

On-farm grass variety evaluation is potentially an alternative means of determining the true agronomic potential of ryegrass varieties and the current study quantified their productivity when exposed to normal

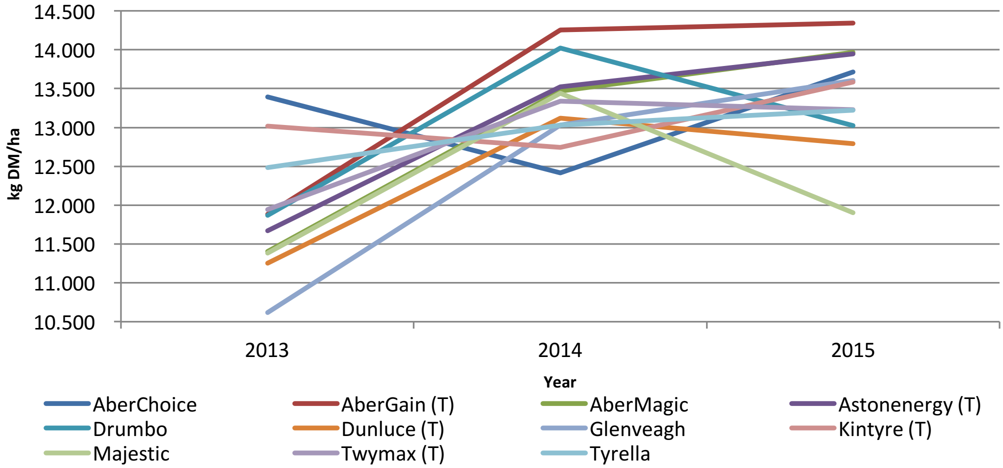

Figure 1. Varietal dry matter production on-farm from 2013-2015.

levels of stress across a range of environments in 'real life' situations. However, the absence of significant differences between varieties was unexpected given that these varieties had all differed in performance when assessed in plot trials. Significant production differences did occur between farms, years and sward age and the levels of variation were not excessive once farm variability was removed. This raises questions regarding the consistency in variety performance between plot trials and farm swards, which needs to be resolved by further investigation. Such additional work is justified as on-farm evaluation has the ability to positively influence both grass breeding objectives toward farm requirements and farmer awareness of the benefits and need to reseed. The current network of on-farm grass growth monitoring through the Irish PBI national grassland database is therefore an important means of continuing this investigation and improving the assessment methods. Ultimately, the knowledge transfer from this initiative is expected to impact on grassland performance potential for ruminant farmers in Ireland.

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Early lactation pasture allowance and duration: the effect on yield of milk fat and protein

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Abstract

Early spring pasture growth can be low, resulting in a pasture availability deficit. The objective of this experiment was to investigate if different pasture allowances offered to early lactation grazing dairy cows for varying time durations influenced yield of milk fat and protein. The two year experiment offered cows one of four pasture allowances (PA; 60, 80, 100 or 120% of intake capacity) for either 2 or 6 weeks. Once the 2- and 6-week time durations had elapsed, the treatments were offered 100% of intake capacity. During the first 2 weeks of the experiment yield of milk fat and protein increased linearly with increasing PA (1.73, 1.77, 1.84 and 1.98 kg cow^{-1} day^{-1}; 60, 80, 100 and 120% PA, respectively). At the end of the 6 weeks there was an effect of the interaction between pasture allowance and duration on yield of milk fat and protein on milk solids yield. All 2-week cows had similar cumulative milk yields for the first 6 weeks of the experiment (78 kg); although they were similar to the 100x6 cows they were different to all other 6-week treatments. The 60 and 80% 6-week cows produced similar yields of milk fat and protein to each other (70 kg), but lower than the 100 and 120% 6-week cows, who differed significantly from each other (80 and 87 kg, respectively). This suggests that in early lactation the effect of PA yield of milk fat and protein depends on the severity and duration of the reduction in feed allowance.

Keywords: pasture allowance, early lactation, dairy cow

Introduction

Low over-winter pasture growth in Ireland (Hurtado-Uria *et al.*, 2013) can result in low pasture availability during early spring, which is a challenge in intensive grazing systems. In anticipation of quota removal in 2015 Irish herd sizes started to increase and it is envisaged that this increase will continue. As land is frequently limited stocking rates on farms have increased, which may further exacerbate the problem of low pasture availability in spring. Restricting pasture allowance (PA), by altering post-grazing height, for a ten-week period in early lactation has previously been shown to reduce immediate milk production but cumulative milk production was unaffected (Ganche *et al.*, 2013). The objective of this experiment was to investigate if different PA offered for varying time durations to grazing dairy cows during early lactation influenced yield of milk fat and protein.

Materials and methods

The study was carried out over a two year period; from March 25 to November 27, 2014 and March 9 to November 23, 2015. Each year 96 dairy cows (41 primiparous and 55 multiparous in year one (Y1); 24 primiparous and 72 multiparous in year two (Y2)) were assigned to a randomised complete block design with a 4×2 factorial arrangement of treatments. Cows were balanced on calving date, breed, lactation number and production variables from the two weeks prior to the start of the experiment. Cows were then randomly assigned to one of four PA (60, 80, 100 or 120% of intake capacity; IC) for either 2 or 6 weeks. Once the 2- and 6-week time durations had elapsed, the treatments were offered 100% of IC. Intake capacity was calculated using the equation of Faverdin *et al.* (2011) based on age, parity, days in milk, bodyweight (BW), body condition score (BCS) and potential milk yield. Pastures were >80% perennial ryegrass. Fresh pasture areas were offered after each milking while treatments were being imposed and on a 24-hour basis thereafter. Pre- and post-grazing sward heights were measured daily using a rising plate meter. Herbage mass (HM; >3.5 cm) was measured twice weekly by cutting 6 strips (120 m^2) per grazing area. Treatment groups grazed adjacent to each other to ensure similar HM was offered. Pasture allowance (>3.5 cm) for the 60, 80 and 120% treatments were calculated based on the IC of the 100×6 treatment. As HM was similar between treatments daily area allocations were different between treatments. Milk yield was recorded daily and milk composition was measured weekly. Data were analysed using covariate analysis and mixed models in SAS v9.3. Terms for year, parity, breed, PA, duration and the interaction of PA and duration were included. Pre-experimental values were used as covariates in the model.

Results and discussion

The mean PA for the 60, 80, 100 and 120% treatments for weeks 1 and 2 were 8.0, 10.7, 13.4 and 16.0 kg DM cow^{-1} day^{-1}, respectively ($P<0.001$). This resulted in post-grazing heights (PGH) of 2.4, 2.9, 3.5 and 4.1 cm, respectively ($P<0.001$). The mean PA and PGH during weeks 3-6 were 9.1, 12.1, 15.1, 18.2 kg DM cow^{-1} day^{-1} and 2.5, 3.1, 3.8, 4.3 cm for the 60, 80, 100 and 120% 6-week treatments. Pasture allowance and PGH for the 2-week treatment, which was grazed as a single herd during weeks 3-6, were 14.8 kg DM ha^{-1} and 3.7 cm, respectively.

There was no effect of duration or breed on yield of milk fat and protein during the first two weeks of the experiment, there was an effect of year as production was higher in the second year of the study due to a more mature herd profile. During the first 2 weeks of the experiment there was a linear increase with increasing PA (1.73, 1.77, 1.84 and 1.98 kg cow^{-1} day^{-1}; 60, 80, 100 and 120% PA, respectively; Table 1).

There was a significant effect of the interaction between PA and duration on yield of milk fat and protein during weeks 3-6 ($P<0.001$). Average daily yield of milk fat and protein during this period was similar for the 2-week treatments (1.86 kg cow^{-1} day^{-1}) indicating no carryover effect of PA offered during the first two weeks of the experiment. Although the two-week treatments were similar to the 100×6 cows they were different to all other 6-week treatments. As expected, because of PA treatments still being imposed, there were differences among the 6-week treatments. The 60×6 and 80×6 treatments had similar yield of milk fat and protein to each other (1.66 kg cow^{-1} day^{-1}), but were lower than the 100×6 (1.91 kg cow^{-1} day^{-1}) and 120×6 (2.08 kg cow^{-1} day^{-1}) treatments, which were significantly different to each other.

There was a significant effect of the interaction between PA and duration on the cumulative yield of milk fat and protein for the first six weeks of the experiment ($P<0.001$). Six week cumulative yield of milk fat and protein was similar for all 2-week treatments (78 kg cow^{-1}) and they were similar to the 100×6 cows (80 kg cow^{-1}) but different to all other six-week treatments. The 60x6 and 80×6 were similar (70 kg cow^{-1}) but lower than the 100×6 and 120×6 (87 kg cow^{-1}), which also differed from each other ($P<0.01$).

Table 1. Yield of milk fat and protein of early lactation dairy cows offered 1 of 4 pasture allowances for either 2 or 6 weeks.[1,2]

	Treatments									Significance			
	60×2	80×2	100×2	120×2	60×6	80×6	100×6	120×6	SED	PA	D	PA×D	Brd
Wks 1 and 2 (kg d^{-1})	1.75	1.78	1.82	1.95	1.72	1.76	1.86	2.02	0.045	0.001	0.624	0.630	0.840
Wks 3 to 6 (kg d^{-1})	1.84^a	1.86^a	1.82^a	1.92^a	1.62^b	1.70^b	1.91^a	2.08^c	0.042	0.001	0.241	0.001	0.557
Cumulative wks 1-6 (kg cow^{-1})	76^a	77^a	77^a	80^a	68^b	71^b	80^a	87^c	1.8	0.001	0.362	0.001	0.670
Cumulative 33 wks (kg cow^{-1})	351^a	338^a	323^a	343^a	329^a	340^a	344^a	374^b	10.3	0.104	0.250	0.064	0.530

[1] Pasture allowance (PA) 60%, 80%, 100% or 120% of intake capacity; D = duration; SED = standard error of the difference; Brd = breed;

[2] Superscripts denote significant differences between treatments with regard to the PA×D interaction.

When cumulative yield of milk fat and protein production for the entire experimental period (33 weeks) was examined there was a tendency (P=0.064) for an interaction between PA and duration. The cows assigned to the 120×6 treatment tended to have a higher yield of milk fat and protein production when compared to all other treatments. No differences were observed between any of the other treatments.

Conclusions

Although early lactation yield of milk fat and protein is reduced when PA is restricted there is little effect when cumulative lactation yield of milk fat and protein is considered compared to cows offered 80% or 100% of IC, however offering 120% of IC for six weeks in early lactation tended to increase yield of milk fat and protein. All other variables such as cow BW and BCS and fertility need to be considered in order to obtain a complete picture of the residual effects of altering PA in early lactation.

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Effects of feeding red clover compared to ryegrass silage to growing cattle out-wintered on kale

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Abstract

Using forage brassicas provides farmers with an alternative to increase the amount of grazed forage in the diets and/or stocking numbers without the need for capital investment in housing. However, typically forage brassicas, such as kale (*Brassica oleracea*), have a low dry matter content and require the addition of a high-fibre source (as hay, straw or silage) to maintain rumen function. Feeding kale at high inclusion rates can reduce intakes due to the low dry matter content of the diet. Feeding ensiled red clover (*Trifolium pratense*) compared to ryegrass (*Lolium perenne*) has been shown to improve the performance of ruminants due to its high protein content and high voluntary intakes compared to ryegrass silages. An experiment was conducted to determine the effects of feeding red clover silage compared to ryegrass silage as the basal high-fibre source to growing beef steers grazing kale over the winter period. Overall, data showed there was no effect of feeding red clover silage compared to ryegrass silage on the performance of beef steers grazing kale.

Keywords: *Brassica oleracea*, forage brassica, *Trifolium pratense*, grazing, steers, ruminants

Introduction

Using forage brassicas allows farmers with an alternative to increase the amount of grazed forage in the diets and/or stocking numbers without the need for capital investment in housing. Out-wintering systems have also been shown to provide benefits to animal welfare and behaviour. Boyle *et al.* (2008) reported significantly more comfort, social and play behaviours and lower limb lesion scores in cattle kept outdoors compared to housed cattle. Other research has shown that the health condition of cows can be positively influenced by outwintering due to the absence of respiratory diseases and infections with ectoparasites (Wassmuth, 2003). However, typically forage brassicas, such as kale (*Brassica oleracea*), have a low dry matter content due to their relatively low neutral detergent fibre (NDF) concentration (Barry and Manley, 1985) and require the addition of a high-fibre source (as hay, straw or silage) to maintain rumen function. Feeding kale at high inclusion rates can reduce intakes due to the low dry matter content of the diet. Feeding ensiled red clover (*Trifolium pratense*) compared to ryegrass (*Lolium perenne*) has been shown to improve the performance of ruminants due to its high protein content and high voluntary intakes compared to ryegrass silages. An experiment was conducted to determine the effects of feeding red clover silage compared to ryegrass silage as the basal high-fibre source to growing beef steers grazing kale over the winter period.

Materials and methods

Six replicated plots (25 m wide × 200 m) of kale (cv. Maris Kestrel) were established in June 2015 by direct drilling into an existing ryegrass sward at the seed rate of 6.9 kg ha^{-1}. Fertiliser was applied in two applications at a total rate of 119 kg N, 19 kg P_2O_5, 67 kg K_2O and 36 kg SO_3 ha^{-1}. Within the width of each plot, there was a 2 m wide strip of existing ryegrass at each edge as lie-back areas. Kale plots were assigned one of two treatments, either ryegrass silage (G) or red clover silage (RC). A total of 36 Aberdeen Angus × dairy steers were allocated to 6 plots of 6 animals based on weight, condition score and age. Prior to the experiment all required vaccines were administered and the steers received an oral flukicide (Fasinex 240, Novartis Animal Health UK Ltd., Basingstoke, UK), an anthelmintic (Ivomec

Super, Merial Animal Health Ltd, Harlow, UK) and a Co, Se, I and Cu bolus (Tracesure, Animax Ltd, Stanton, UK).

On 8 October the cattle were weighed (mean live weight (LW) 424 kg, standard deviation (sd) 25.4 kg) and moved from a predominantly ryegrass pasture onto their respective plots with access to grazed grass, kale and their allocated silage diet. The eight week measurement period started on 15 October, following adaptation to diet and the steers were weighed weekly.

Each week and within each plot, triplicate 1 m^2 quadrats of kale in the area to be grazed during the coming week were harvested to ground level. In addition, triplicate 1 m^2 quadrats from the area grazed during the preceding week were harvested to ground level to determine residual kale yield. Post-grazing samples were washed to remove soil and stone and drained overnight at 4°C before weighing. Electric fences were used to allocate an area of kale estimated to provide 0.7 of total dry matter (DM) intake (assuming 80% utilization of the kale crop) and prevent grazing on any regrowth. Fences were moved on a daily basis and water troughs and silage feeders were moved regularly as the grazing areas moved along the plots.

In June 2015, first cut red clover (cv. Milvus) and first cut permanent grass pasture silage was mown and ensiled in round bales without an additive and following a 48 h wilt. Chopped silage was offered to each steer group based on their LW with a daily DM allocation of 0.3 (0.024 LW) designed to achieve 0.3 of total DM intake as silage. Dry matter content of the kale, silages and any refusals was determined by drying at 100 °C for 48 h. Daily sub-samples of silages offered were bulked weekly for DM determination at 100 °C and further samples were freeze dried prior to determination of nitrogen (N) and pH was determined on fresh frozen silage samples.

Weekly kale intake was estimated by subtracting post-grazing yield from pre-grazing yield and multiplying by the total area of kale grazed. Intakes adjusted for LW were analysed by repeated measures ANOVA. Individual steer LW gains were calculated using the non-parametric regression method of Theil (1950) and were analysed by one-way ANOVA using individual LW on day -7 as a covariate and with kale plot as the experimental unit.

Results and discussion

The mean DM of the ryegrass and red clover silage as offered over the experimental period was 514 (sd 25.9) and 434 (sd 21.7) g kg^{-1} fresh weight, respectively. The mean N concentration of the ryegrass and red clover silage was typical at 14.7 (sd 0.8) and 25.6 (sd 0.3) g kg^{-1} DM, respectively. The mean pH of the ryegrass and red clover silages were 4.7 (sd 0.1) and 4.4 (sd 0.1), respectively, indicating that both silages were well preserved. Mean kale availability was 8.7 tonnes DM ha^{-1} with a mean DM content of 123 g kg^{-1}. Approximately 19% (range 14 to 30) was unutilised each week. Estimated kale intake showed no difference between silages (Table 1) and equated to approximately 7.3 kg DM per head per day and 70% of total DM intake. These data confirm that the voluntary intake of kale did not differ between treatments, that there was no effect of the kale on voluntary intake of the cattle, as seen in studies offering cattle 100% kale diets (Keogh *et al.*, 2009), and that the experiment reported here compared differences in the silage treatment at 30% of total DM intake. There were no difference in the liveweight gain of beef steers offered either red clover or ryegrass silage in addition to fresh kale forage. Previous studies on kale offered to young cattle found only liveweight gains on kale of between 250 and 425 g d^{-1}, the latter with copper supplementation (Barry *et al.*, 1981) or rates of daily gain of steers offered proportions of 0.50 or 0.75 kale with rolled barley or 100 kale were 1.05, 0.75 and 0.76 kg d^{-1}, respectively (Macdearmid *et al.*, 1982). Therefore, the high liveweight gain observed in the current experiment overall may be one explanation for the lack of treatment effect found.

Table 1. Intake and liveweight (LW) gain by steers grazing kale and offered either ryegrass (G) or red clover silage (RC).

	Silage		sed	Prob
	G	RC		
Intake (g DM kg LW^{-1} d^{-1})				
Total	22.7	22.5	0.25	0.616
Silage	6.9	7.1	0.12	0.255
Kale	15.8	15.5	0.15	0.112
LW gain (kg $head^{-1}$ d^{-1})	0.907	0.937	0.0817	0.717

Conclusions

Grazing beef steers on kale gained on average over 900 g d^{-1}. Overall, for beef steers consuming 70% of their DM intake as kale, there was no effect feeding red clover silage compared to ryegrass silage on their intake or performance of beef steers grazing kale.

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The Moorepark grass growth model: application in grazing systems

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Abstract

The Moorepark Grass Growth model (MGGm) is a dynamic model developed in C++ describing the grass biomass evolution of a paddock over time with a daily time step. It is assumed that each urine and faeces deposition affects 2 m^2 area of the paddock, with the model describing the different variables and events such as grass growth, mineralisation, immobilisation, leaching and nitrogen (N) uptake by the plant at the 2 m^2 level. Different N mineral fertilisations in a grazing context have been simulated, and the impact on grass growth and N content of the grass has been evaluated. The grazing simulations examined, described the effect of N returns on grass growth and N fluxes accurately. The model has been able to react to different weather conditions, and to different levels of N mineral fertilisation. The response of the grass growth, the biomass produced and the grass N content to the N mineral fertilisation level is in line with the literature. In average, the mineral N fertilisation leaded to an increase of grass utilised of 12 and 17 kg of dry matter per kg of N fertilised for the French and Irish simulation respectively.

Keywords: grass growth, model, grazing, dairy cow

Introduction

In temperate climates grass provides the cheapest and highest quality feed for dairy cows (Dillon *et al.*, 2005). However, even though a temperate climate allows grass growth throughout the year, grass growth is highly seasonal and depends heavily on climate conditions. Management of pasture (such as fertilisation, cutting and grazing heights) is also an important factor influencing grass growth. There is increased interest in the potential to increase grass growth and utilisation through more advanced grassland management. This paper presents the prediction of the Moorepark Grass Growth model (MGGm) in terms of grass growth and nitrogen content of the grass in a grazing context with different mineral nitrogen fertilisation levels and different weather conditions.

Materials and methods

The MGGm is a dynamic model developed in C++ describing the grass growth and the N fluxes of a paddock at a 2 m^2 level. The model is run with a daily time step simulating soil N mineralisation/ immobilisation and water fluxes, grass growth, N uptake and grass N content. The model is driven by a daily potential growth depending on the radiation and the total green biomass. To calculate the actual daily growth, this potential growth is then multiplied by parameters depending on environmental conditions (temperature, water in the soil and radiation) and a parameter depending on the availability of the mineral N in the soil compared to the demand in N associated with the potential grass growth. The availability of the N in the soil depends on the mineral N in the soil, the proportion of the N usable by the plant (depending on the time of the year and the heading date) and the N demand to grow one kg of biomass (depending on the N dilution curve and the actual biomass over 4 cm; Gastal and Lemaire, 2002). The N dilution curve represents the decrease of the N needed for one kg of dry matter (DM) growth with the increase of the accumulated biomass (Gastal and Lemaire, 2002). Animal deposition of nitrogen is simulated through a one day-grazing sequence, with the number of animals adapted by the

Table 1. Description of the two annual weathers applied in the simulations.

Location	Annual average temperature (°C)	Monthly rainfall (mm)												Annual total rainfall (mm)
		J	F	M	A	M	J	J	A	S	O	N	D	
France Normandie	10.2	61	32	136	68	64	12	35	72	42	80	102	41	743
Ireland Co. Cork	8.7	107	39	88	59	38	53	143	23	102	83	98	37	869

model depending on the pre grazing mass (calculated by the model) based on a daily intake of the animal which was assumed to be 16 kg DM ha^{-1}.

Different simulations have been run to simulate the impact of mineral N fertilisation in grazing conditions. These simulations have been tested on 2 different weather conditions (Table 1) in Ireland (Co. Cork) and in France (Normandy). The initial mineral N in the soil was defined at 80 and the organic N at 14,400 kg N ha^{-1} (6% of organic matter content). Four different mineral N applications were tested 0, 100, 200 or 300 kg of N ha^{-1} per year in Ireland. Due to the lack of rain and high temperature in summer in Normandy, the annual mineral N fertilization has been reduced by 25% without applications in autumn. The size of the paddock is 1 ha and a total of seven (France) or eight (Ireland) grazing events has been conducted.

Results and discussion

The results are presented in Table 2 and Figure 1. The model is capable of reacting to different weather conditions and is highly sensitive to different levels of mineral N fertilisation (Figure 1). The lack in rainfall in France in 2008 led to a poor growth in summer and autumn. The response to N mineral level of fertilisation conforms with the literature (Whitehead, 1995) with a higher response at low mineral N levels than at higher applied levels (20.2 and 14.8 kg DM kg^{-1} N applied between 0 to100 or 75 kg N and 13.1 and 8.9 kg DM kg^{-1} N mineral applied between 200 and 300 or 150 and 225 kg mineral N respectively in Ireland and in France).

The N content of the grass offered evolves in accordance with the season and increases with the mineral N fertilisation levels. This higher concentration has been previously described in the literature (Whitehead, 1995). When there is higher mineral N available in the soil there is higher N uptake. This is in accordance with the principle of the dilution curve described by Gastal and Lemaire (2002). Due to the weather conditions in France, the annual average N content of the grass offered is a little bit higher than in Ireland. This is because in autumn, the high level of soil N mineralisation is accompanied by a lower grass growth than in Ireland. Consequently the relative N uptake is higher leading to a higher N concentration in the plant.

Table 2. Impact of the weather conditions and the mineral N fertilisation on the total grass grazed and average grass N content offered at grazing.[1]

Mineral N fertilisation (France / Ireland)		0	75 / 100	150 / 200	225 / 300
Total grass grazed (kg DM ha^{-1})	France, Normandie	6,014	7,151	8,023	8,710
	Ireland, Co. Cork	8,388	10,446	12,120	13,452
Average grass N content offered	France, Normandie	2.52	2.73	2.94	3.14
(g kg^{-1} DM)	Ireland, Co. Cork	2.30	2.56	2.81	3.04

[1] DM = dry matter.

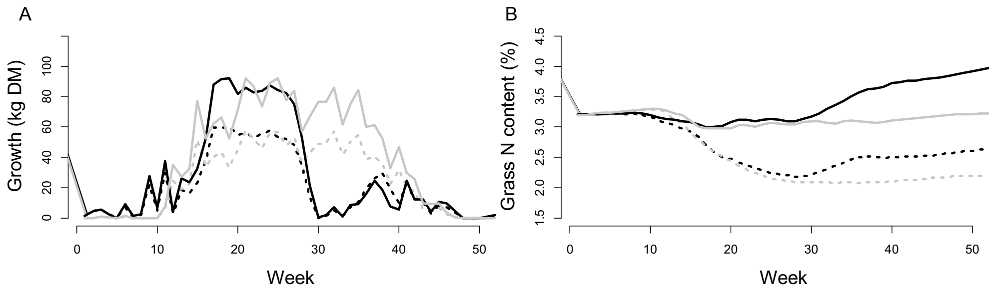

Figure 1. Net growth and grass N content for the French 2008 weather (black) and the Irish 2009 weather (grey) at the level 0 (dotted line) and 300 (continuous line) of mineral N fertilisation.

Conclusions and further work

The MGGm is a user friendly model with basic and simple inputs. The model has been capable of reacting in a sensible manner to different weather and mineral N fertilisation events. The future of the MGGm is to be included in a whole farm dairy model (the Pasture Base Herd Dynamic Milk model (Ruelle *et al.*, 2015)), permitting the prediction of the impact of different climatic conditions, grazing management and fertilisation practices on the milk production of the animals and the economics of the farm.

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Effect of ewe prolificacy potential as dictated by sire breed and stocking rate on grass utilisation and lamb carcass output

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Abstract

Grazed grass is the most economic source of nutrition available to ruminants in temperate grazing regions and has the potential to supply up to 95% of the energy requirements of sheep. This study investigated the effect of ewe prolificacy potential (PP) and stocking rate (SR) and their interaction on herbage utilisation and lamb carcass output (kg ha^{-1}) from a grass-based lamb production system. The study was carried out over two production years and had a 2×3 factorial design, consisting of two PP levels as dictated by sire breed and three SR's incorporating 360 ewes (180 medium prolificacy potential (MP – Suffolk crossbred ewes; target weaning rate 1.5 lambs per ewe) and 180 high prolificacy potential ewes (HP – Belclare crossbred ewes; target weaning rate 1.8 lambs per ewe)). Stocking rates were low (LSR; 10 ewes ha^{-1}), medium (MSR; 12 ewes ha^{-1}), and high (HSR: 14 ewes ha^{-1}). As SR increased the quantity of grass utilised per hectare (P<0.001), per ewe and lamb unit (ewe plus lambs reared; P<0.05) and per kg of lamb carcass produced (P<0.01) increased. Carcass output per hectare increased as both SR (P<0.001) and PP (P<0.01) increased. This study shows the potential to increase carcass output from a grass based system through greater PP and SR levels.

Keywords: stocking rate, prolificacy, herbage utilisation, carcass output

Introduction

Maximising the quantity of herbage utilised and the proportion of carcass produced from a grass based diet has the potential to greatly increase farm profitability as grazed grass is the cheapest source of nutrition available to grazing ruminants (Finneran *et al.*, 2010) and has the potential to supply up to 95% of the energy requirements of sheep (Davies and Penning, 1996). This provides lamb producers in temperate grazing regions the opportunity to produce and finish lambs from a grass based diet. Therefore the objective of this study was to investigate the effect of ewe prolificacy potential (PP; predicted number of lambs born per ewe per year) as dictated by sire breed and stocking rate (SR; ewes ha^{-1}) and their interaction on the level of grass utilised per hectare, per ewe and unit (ewe plus lambs reared), per kilogram of carcass produced and on carcass output (kg ha^{-1} and kg ewe^{-1}) from a grass-based lamb production system.

Materials and methods

The study was conducted at the Sheep research demonstration farm, Teagasc, Animal & Grassland Research and Innovation Centre, Athenry, Co, Galway, Ireland (54°80'N; 7°25'W), from October 2012 for two production years. The study had a 2×3 factorial design which consisted of two PP levels as dictated by sire breed. This comprised of 180 medium prolificacy (MP) – Suffolk crossbred ewes (target weaning rate of 1.5 lambs per ewe), and 180 high prolificacy (HP) – Belclare crossbred ewes (target weaning rate of 1.8 lambs per ewe; Hanrahan, 1994). Within PP, ewes were blocked by live weight (kg) and body condition score (BCS) on a scale of 1 to 5, before being randomly assigned to one of three SR pasture systems: a low (LSR; 10 ewes ha^{-1}), a medium (MSR; 12 ewes ha^{-1}) or high (HSR; 14 ewes ha^{-1}) in October 2012. Ewes were mated to Charollais rams over a six week period in October and November each year. Post-lambing, ewes and their lambs were turned out to pasture and grazed in a five paddock rotational system to post-grazing sward height targets of 4.5, 4.1, and 3.7 cm for the LSR, MSR and HSR, respectively. Lambs were weaned on average at 14 weeks of age, with a leader follower grazing system operated post-weaning. Lambs were drafted for slaughter once pre-defined live weight targets of 42, 43, 44, 45 and 46 kg were reached in the months June, July, August, September and October, respectively to produce a carcass of 20 kg. Nitrogen fertiliser was applied at a rate of 13 kg ewe^{-1} across all treatments. Pre- and post-grazing (>3.5 cm) herbage mass (kg dry matter (DM) ha^{-1}) was determined for each grazing by harvesting two random strips (1.2×10 m) of grass representative of the paddock with an Etesia mower. All mown herbage from the strip was collected, weighed and a 0.1 kg (fresh weight) sub sample taken and dried for 16 hours at 90 °C for the calculation of DM content and herbage quality. Lambs that were not drafted by October in each treatment were removed from their grazing farmlet when grass supply dropped below 50 kg DM per ewe per ha or lamb growth rate dropped below 100 grams per day and finished indoors on grass silage *ad libitum* and concentrates. Lamb carcass output per ha produced from grazed grass alone and total carcass output (includes lambs finished on grass silage and concentrates) was recorded. The effect of PP, SR and their interaction on herbage utilisation and carcass output were modelled using linear mixed models in PROC HPMIXED (SAS Inst. Inc., Cary, NC, USA).

Results and discussion

The interaction between PP and SR was tested but was not significant and is not reported. The number of lambs weaned per ewe ($P<0.01$) was 1.49 (MP) and 1.70 (HP), respectively. Increasing SR significantly increased the quantity of herbage utilised per hectare ($P<0.001$; Table 1) and supports previous findings in a stocking rate study using dairy cows by MacDonald, *et al.* (2008) of increased herbage consumption as SR increases. Herbage utilised per ewe and lamb unit and per kilogram of carcass produced increased as SR increased ($P<0.01$), although the LSR and MSR systems did not differ from each other and had a lower carcass output on a per hectare basis relative to the HSR system ($P<0.01$). Ewe PP did not affect the quantity of herbage utilised per ewe and lamb unit, indicating both the MP and HP systems had similar feed utilisation levels. Carcass output per hectare increased as SR and PP increased ($P<0.001$). On a ewe basis SR had no effect on carcass output while ewe prolificacy potential did, with HP ewes

Table 1. The effect of ewe prolificacy potential (PP)[1] and stocking rate (SR)[2] on the quantity of herbage utilised per hectare, per ewe and lamb unit (ewe plus lambs reared) and per kilogram of carcass and carcass output.[3]

	PP			SR				*P*-value	
Parameter	MP	HP	SEM	LSR	MSR	HSR	SEM	PP	SR
Kilograms of grass dry matter utilised									
Per hectare	9 839	9 974	39.6	7 999^a	9 472^b	12 226^c	49.0	NS	***
Per ewe and lamb unit	815	827	4.1	799^a	789^a	875^b	4.9	NS	**
Per kilogram of carcass	26.2	26.6	0.13	24.9^a	25.0^a	29.3^b	0.15	NS	**
Total kilograms of carcass produced from grazed grass (kg)									
Per hectare	289	350	6.9	297^a	320^a	342^b	8.6	***	***
Per ewe	24.6	29.4	0.55	29.7^a	26.7^b	24.5^b	0.67	**	**
Total kilograms of carcass produced (kg)									
Per hectare	344	401	7.3	320^a	379^b	419^c	8.9	***	***
Per ewe	28.8	33.5	0.56	32.0	31.6	30.1	0.69	**	NS
Prop. of carcass output from grass[4]	0.85	0.88	0.01	0.93^a	0.84^b	0.81^b		NS	**

[1] Prolificacy potential: MP = medium prolificacy (target weaning rate = 1.5 lambs per ewe), HP = high prolificacy (target weaning rate = 1.8 lambs per ewe).

[2] Stocking rate: LSR = 10 ewes ha^{-1}; MSR = 12 ewes ha^{-1}; HSR = 14 ewes ha^{-1}. Number of animals used = 360 ewes and 1,148 lambs; SEM = standard error of the mean.

[3] Within rows superscripts indicates significant difference; * = $P<0.05$, ** = $P<0.01$, *** = $P<0.001$, NS = $P>0.05$.

[4] Proportion of carcass output from grazed grass only (calculated as the ratio of carcass (kg) from grass only relative to total carcass output (kg)).

producing 4.7 kg more carcass on a per ewe basis relative to MP ewes (P<0.01) as a result of the greater number of lambs reared per ewe. The proportion of carcass produced from grazed grass alone was greatest at the LSR, intermediate at the MSR and lowest at the HSR, however the MSR and HSR systems did not significantly differ from each other (0.93, 0.84 and 0.81; P<0.01).

Conclusions

Increasing SR increased the quantity of grass utilised per hectare, per ewe and lamb unit and per kilogram of lamb carcass produced. Greater carcass output on a per hectare basis is achievable through increasing PP and SR, although there was a limit in terms of efficiency of turning grass into carcass output, with similar proportions of carcass produced from grazed grass in the 12 and 14 ewes per hectare systems.

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The timing of access to pasture affects ingestive behaviour and milk yield of dairy ewes

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Abstract

The effects of the timing of access to pasture (AM, from 08:00 to 12:00; PM, from 15:30 to 19:30) on ingestive behaviour and performance were assessed in dairy ewes rotationally grazing Italian ryegrass in spring. A randomised block design with two replicates per treatment was used. Twenty-four mid lactation ewes were divided into 4 six-ewe groups, homogeneous for milk yield and body weight and randomly allocated to the treatments. The ewes were supplemented daily with 700 g head^{-1} of concentrates and 700 g head^{-1} of a ryegrass-based hay. Pasture plot and animal group data were analysed by a factorial model including timing of access to pasture, grazing week and their interaction as fixed effects. The effect of experimental week was significant on all the studied variables (P<0.05), whereas the interaction was not (P>0.05). Sward height and herbage mass did not differ between treatments (P>0.05), averaging 20.1 cm and 2.33 t DM ha^{-1}, respectively. Dry matter and water soluble carbohydrates were higher in herbage hand-plucked samples collected from PM than AM subplots (P<0.01). Grazing time (P<0.01) and herbage intake (P<0.05) were higher in PM than AM ewes. Fat normalized milk yield was also the highest in PM ewes (P<0.05). To conclude, giving access to pasture in the afternoon rather than in the morning increased grazing time, herbage intake and milk performance of ewes grazing Italian ryegrass in spring.

Keywords: sheep, nutrition, grazing management, chronophysiology

Introduction

The timing of access to pasture has been shown to impact on feeding behaviour, intake and performance of ruminants, as reviewed by Gregorini (2012). In particular, the offer of pasture during afternoon rather than morning hours can enhance bite mass, bite rate and hence intake rate of cattle and increase growth rate of beef cattle and milk yield in dairy cows, particularly in early lactation. In small ruminants, which are more selective than cattle, data are still scanty. Thus, the objective of this study was to evaluate the effect of the timing of access to ryegrass pasture in spring on ingestive behaviour and milk performance of dairy sheep.

Materials and methods

The experiment was run at Bonassai research station (NW Sardinia, Italy). One hectare of Italian ryegrass (*Lolium multiflorum* Lam., cv. Teanna) was sown in January (sowing rate of 40 kg ha^{-1}) and fertilized with 46 kg ha^{-1} of P_2O_5 and 64 kg ha^{-1} of N. The experiment lasted seven weeks in total from March to May 2015, with six experimental weeks. A randomized block design with two replicates was adopted. Two blocks of 5,000 m^2 were divided into two plots of 2,500 m^2 each. Each plot was then split into four subplots, rotationally grazed with a grazing cycle of 7 days and a regrowth period of 21 days. Treatments were two different times of access to pasture: morning (AM, from 08:00 to 12:00) and afternoon (PM, from 15:30 to 19:30). The plots were randomly assigned to the treatments within each block.

Twenty-four mature Sarda ewes in mid-lactation (mean ± standard deviation, 92±3 days in milk) were used. Based on pre-experimental milk yield (1,712±239 g milk ewe^{-1} day^{-1}), live weight (40.6±3.4 kg) and body condition score (2.47±0.2 units), they were divided into four groups of six ewes, homogeneous for the above criteria. The groups where then randomly assigned to the treatment plots. The ewes were machine-milked twice daily at 7:00 and 15:00. Each day the ewes were carted to the corresponding subplots where stayed for the planned time. During the remaining day-time the ewes were housed, receiving a daily supplementation of 400 g head^{-1} of a commercial concentrate split into two equal meals at milkings, and 300 g head^{-1} of maize grain and 700 g head^{-1} of a ryegrass-based hay, both supplied at pasture turn-out.

Sward height was measured using a weighted-plate grass-meter (50 random measurements per subplot on three occasions during each grazing cycle). Herbage mass was determined at the beginning of each grazing cycle by cutting four quadrats of 0.5 m^2 per subplot at 3 cm above ground level. Herbage composition and animal measurements were taken on the mid-day of the six grazing cycles (test day). Herbage samples were hand-plucked from each grazed subplot at 10:00 (subplots AM) or 17:30 (subplots PM), oven-dried at 65 °C for 72 h and then ground to determine the content of dry matter (DM), crude protein (CP), neutral detergent fibre (AOAC, 1990), and water soluble carbohydrates (WSC; Deriaz, 1961). Short-term intake rate was measured on five ewes per group using the double-weighing technique (Penning and Hooper, 1985). Briefly, herbage intake rate (g min^{-1} grazing) was measured weighing the ewes before and after the first hour of access to pasture on a precise electronic scale (Multirange, Mettler Toledo, Novate Milanese, Italy). An additional ewe per group, which rotated among animals within group on each test day, was used to simultaneously estimate insensible weight losses (IWL). On each test day, the intake rate of each group was corrected for the IWL of the ewe of the same group. The grazing time (GT) was measured by direct observation of the same five animals used for intake rate measurements every 3 min throughout the grazing sessions. Herbage intake was then computed by multiplying the herbage intake rate (not shown) of each ewe by the corresponding GT. The day after each test day, milk yield of the ewes tested for intake was measured and milk samples assayed for fat content (Milkoscan FT+, Foss Electric, Hillerød, Denmark). Milk yield normalised at 6.5% fat was then computed (Pulina *et al.*, 1989).

Average pasture subplot and animal group data were analysed by analysis of variance using a model inclusive of the fixed effects of treatment, week and their interaction. The interaction was not significant for any variable under study.

Results and discussion

Sward height and herbage mass did not differ between treatments (P>0.05) averaging 20.1 cm and 2.33 t DM ha^{-1}, respectively, whereas they varied markedly throughout the experiment (week effect: P<0.05) due to the physiological development of Italian ryegrass. The week effect was also overriding on all the herbage and animal variables under study (P<0.05). The pasture in the afternoon had higher content of DM and WSC, the latter causing, probably for a dilution effect (Gregorini, 2012), a decrease of content of ash and CP (Table 1). Grazing time and herbage DM intake were higher in PM than AM dairy ewes, as found by Avondo *et al.* (2008) in dairy goats grazing berseem clover. Milk yield and fat normalized milk yield were also higher in PM ewes. The slight increase (c.a. 5%) is consistent with findings in cattle, such as those by Kennedy *et al.* (2009).

Table 1. Effects of timing of access to pasture (TRT: from 8:00 to 12:00 (AM) or from 15:30 to 19:30 (PM)) and experimental week (WK) on hand-plucked herbage composition, grazing time, herbage intake and milk production of dairy ewes rotationally stocked on Italian ryegrass in spring.[1]

	Treatment means			Sig. of Probability <[2]	
	AM	PM	RMSE	TRT	WK
Herbage composition					
DM (% as fed)	19.50	21.51	1.68	0.001	0.01
Ash (% DM)	11.82	10.66	1.03	0.05	0.05
NDF (% DM)	37.19	35.51	3.03	ns	0.001
CP (% DM)	16.53	15.10	1.01	0.01	0.001
WSC (% DM)	20.53	24.33	0.73	0.01	0.001
Grazing time (min/day^{-1})	187.9	205.5	11.0	0.001	ns
Herbage intake (g DM ewe^{-1} day^{-1})	1,312	1,479	188	0.05	0.05
Milk yield (g ewe^{-1} day^{-1})	1,410	1,473	73.4	0.05	0.01
Fat normalized MY (g DM ewe^{-1} day^{-1})	1,335	1,408	80.1	0.05	0.01

[1] DM = dry matter; NDF = neutral detergent fibre; CP = crude protein; WSC = water soluble carbohydrates; MY = milk yield; RMSE =root of the mean square error.
[2] ns = P>0.05.

Conclusions

To conclude, providing access to ryegrass pasture in the afternoon rather than in the morning increased grazing time, herbage intake and milk performance of grazing ewes in spring.

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Sainfoin accessions exhibit a marked potential for optimisation of proanthocyanidins in the forage

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Abstract

Proanthocyanidins (PAs) have been shown to be useful for improving the sustainability of livestock farming: they reduce methane and nitrous oxide emissions from ruminants, as well as the requirement for drug administration due to their anthelmintic properties. To test the possibility of improving the bioactivity of the forage legume sainfoin (*Onobrychis viciifolia* Scop.), individual plants of 27 sainfoin accessions were analysed for PA concentration and composition. Large variabilities – both among and within accessions – in PA concentrations (23.0-47.5 mg g^{-1} leaf dry matter (DM)), in the share of prodelphinidins (78.6-97.5%), and in the mean degree of polymerization (11-14) were found. This variation provides opportunities for improvement of the concentration and composition of PA in sainfoin. The results also suggest that the speed of plant development should be considered in further selection programmes.

Keywords: *Onobrychis viciifolia*, sainfoin, proanthocyanidins, tannins, variability, accessions

Introduction

Proanthocyanidins (PAs) in specific forage plants can contribute in several ways to increase the sustainability of livestock farming: they prevent bloat, help reduce methane and nitrous oxide emissions, improve utilisation of forage proteins in the animal rumen and are known to have an anthelmintic effect (reviewed by Lüscher *et al.*, 2014). This seems especially to be the case for PAs with a high share of prodelphinidins (PD%) (Mechineni *et al.*, 2014), which emphasises the importance of the composition of the PA. Although sainfoin (*Onobrychis viciifolia* Scop.) is considered to be PA-rich (Häring *et al.*, 2007), both concentration and composition of its PAs require improvement. For effective plant breeding, a wide variability within the genetic resources is important (Boller *et al.*, 2010). To test the possibility for optimisation of bioactivity by plant breeding, 27 sainfoin accessions were screened for variability in both PA concentration and composition.

Materials and methods

A field experiment was established in Rümlang, Switzerland (47°44' N 8°53' E, 482 m a.s.l.) in summer 2012. Seeds of 27 sainfoin accessions of various origins and cultivation status were sown in rows of 9 plants at a spacing of 0.50×0.25 m (between×within). The design was a randomized complete block design with 8 blocks. From sowing to harvest after 126 growing days, plant development was recorded on nine occasions. The speed of plant development was calculated as the sum of stage days, based on a scale of 13 stages. Fully developed leaves were harvested, immediately cooled and stored at -70 °C until freeze drying. Preparation for analysis was conducted as described in Malisch *et al.* (2015). Phenolic compounds were analysed in a UPLC-MS/MS chain and the mean degree of polymerisation (mDP) was calculated according to the methods of Engström *et al.* (2014). The primary response variables PA concentration, mDP and PD% were analysed with a linear-mixed regression model. Accession and development speed of each individual plant were taken as fixed, and block as random, respectively. The data of PD% were logit-transformed prior to statistical analysis. All calculations were conducted with the statistical software R (R Core Team, 2015).

Results and discussion

The means of PA concentration in the leaves among the tested accessions varied significantly ($P<0.001$), covering a range from 23.0 mg PA g^{-1} dry matter (DM) for 'Perdix' to 47.5 for 'WKT 10' (Figure 1A). The within-accession variability was also wide. While 'Perdix' exhibited a standard deviation (sd) of only 3.1 mg PA g^{-1} DM, others as 'CPI 63825', 'TU86-43-03' or '247' varied roughly three-times as much (sd between 8.5 and 9.3 mg PA g^{-1} DM). The low within-accession variability for PAs of 'Perdix' could be regarded as a consequence of its cultivation status as a cultivar (Malisch *et al.*, 2015), and its low average PA concentration is explained because high yields have hitherto been of significant importance in official variety testing rather than this trait (Frick *et al.*, 2011). Interestingly, while the relationship between the development speed of the individual plants of the experiment and their PA concentration was slightly negative ($P<0.001$), it was positive within 'WKT10', 'CPI 63780 and 'Esparsette' which may also suggest further variability.

For the mDP, significant ($P<0.001$) differences between the tested accessions were found (Fig. 1B), ranging from 10.64 for accession 'CPI 63820' to 13.92 for accession '274'. This property of the PAs

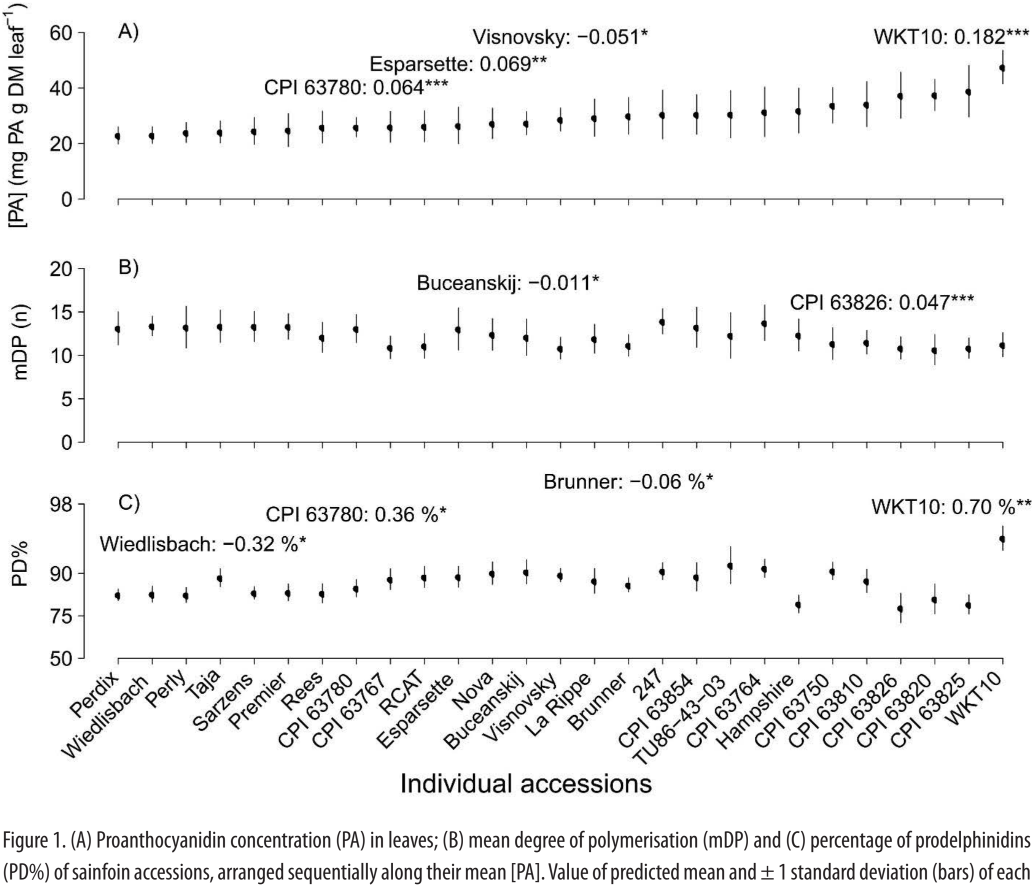

Figure 1. (A) Proanthocyanidin concentration (PA) in leaves; (B) mean degree of polymerisation (mDP) and (C) percentage of prodelphinidins (PD%) of sainfoin accessions, arranged sequentially along their mean [PA]. Value of predicted mean and ± 1 standard deviation (bars) of each accession based on regression analysis is given. Numerical values with accession names indicate in (A) and (B) the coefficient of regression and in (C), due to back transformation, percent change, respectively, dependent on plant development speed. Significance: **P*≤0.05, ***P*≤0.01, ****P*≤0.001.

showed a larger variability within accessions than among them, illustrated by 'TU86-43-03' which exhibited an sd for the mDP of 2.61. This means that about one third of the individuals had an mDP of more than 14.91 (mean + 1 sd) and another third less than 9.69 (mean – 1 sd), resulting in a difference of 5.22 units of polymerisation. The mDP increased with the development speed of the individual plants in the experiment (P<0.001). The accessions 'CPI 63826' and 'Buceanskij' showed differences in this relationship: while in 'CPI 63826' this was manifested by a larger coefficient, the mDP in 'Buceanskij' decreased with increasing plant development speed of its individuals (negative coefficient).

The PD% was also highly variable, both among (P<0.001) and within accessions (Figure 1C). In 'WKT10' the mean PD% was 97.5%, whereas the PAs of 'CPI 63826' consisted of only 78.6% of PD. The maximum difference within the accessions of 12.9% was found in 'TU-86-43-03'. For 'WKT10', the difference in PD% between the upper and the lower third of individuals was 2.6% and, thus, the least of all accessions. Nevertheless, the PD% of 'WKT10' showed the strongest positive response to development speed. This was contrasted by the strongly negative response of PD% to development speed within 'Wiedlisbach', revealing an additional source of variability.

Conclusions

Sainfoin exhibits a marked degree of variability in important properties of proanthocyanidins, both among and within accessions, providing opportunities for improvement by plant breeding. The results suggest that plant development speed should also be considered in further selection programmes.

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The effects of legume-grass inoculation on silage fermentation, aerobic stability and milk yield

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Abstract

A silage inoculant, blend of bacteria strains *Enterococcus faecium, Lactobacillus brevis* and *Lactobacillus plantarum*, was used for legume-grass silage vs an untreated control silage. Mixed grass-legume sward (35% *Lolium perenne*, 15% *Phleum pratense*, 45% *Trifolium pratense* and 5% others species in a second cut) were wilted for 6-8 hours to 32% dry matter chopped and then ensiled into two ferro-concrete trenches (100 t each). Twenty-four dairy cows divided into two analogous groups were fed control and inoculated silages, and feed intake and their productivity was evaluated over a 92 day feeding period. Addition of lactic acid bacteria improved the fermentation profile by lowering pH, butyric acid, ammonia-N and dry matter loss. Treated silage had a higher concentration of lactic and acetic acid and better aerobic stability. Inoculation had small effect on silage dry matter intake or milk yield but utilization of feed energy and the milk hygienic quality was better with inoculated silage.

Keywords: grass, inoculant, milk, production, silage

Introduction

For the balance of the seasonal deficit of feed, the farmer must preserve part of the grass produced in favourable vegetation season in order to guarantee stable animal feeding throughout whole year. Ensiling is an old preservation method for the most forage crops based on lactic acid bacteria activity. It is possible to apply bacterial inoculants at ensiling to promote improved fermentation and to achieve other collateral effects, such as improved aerobic stability (Broberg *et al.*, 2007; Saarisalo *et al.*, 2007). The aim of the trial was to study the effect of the inoculant, blend of *Enterococcus faecium, Lactobacillus brevis* and *Lactobacillus plantarum*, on the fermentation and aerobic stability of first cut grass-legume silage, as well as the effect of inoculation on the feed intake and milk production of dairy cows.

Materials and methods

A mixed grass-legume sward (35% *Lolium perenne,* 15% *Phleum pratense*, 45% *Trifolium pratense* and 5% others, second cut) was wilted to 32% dry matter, chopped and ensiled into two ferro-concrete trenches (100 t each). The following treatments were applied to fresh forage: control (C) had no additive and whereas (T) was inoculated with a silage inoculant, containing *Enterococcus faecium, Lactobacillus brevis* and *Lactobacillus plantarum*. The inoculant suspension was applied via a sprayer mounted on the chopper at rate of 4 l per tonne freshly chopped forage in order to obtain 2×10^5 colony forming units per gram of forage. Five control bags (made from four layers cheesecloth) filled with 1 kg ensiling material were putted in each silo to determine dry matter losses. Forage and silages samples were taken for chemical analyses. Silages also were analysed for aerobic stability by placing thermocouple wires at the centre of bag containing 1 kg silage, within an open top polystyrene box.

Twenty-four milking dairy cows were selected for the feeding experiment. During a three week pre-experimental period, the animals were fed with untreated (control) silage. At the start of the experimental period lasted 92 days, the animals were randomly allocated to two analogous groups of twelve and one group was fed with the control silage when other group with inoculated silage. Equal amounts of concentrate feed were supplemented to each group. The daily silage intake was recorded once a week over two consecutive days and their milk yield was registered once every week. Once a week milk samples were taken from the morning and evening milking and the fat, protein, lactose contents and the count of somatic cells were analysed.

Results and discussion

Application of the inoculant resulted in a significantly lower pH value and in a significantly higher lactic acid concentration compared to the control silage. The concentration of acetic acid was numerically higher in inoculated silage than in not treated (Table 1). This reflects the results obtained by Muck *et al.* (2007).

Furthermore, inoculated silage showed 10 times (P<0.05) lower concentration of the butyric acid and lower by 19.2% (P<0.05) content of ammonia-N. The results may indicate that the extent of proteolysis can be reduced by an application of selective mixtures of lactic acid bacteria. Dry matter loss was significantly lower and net energy lactation (NE_L) was by 1.25% higher (P<0.05) for inoculant treated silage when compare to control silage. Inoculation had a positive effect on aerobic stability of the silage. The temperature rise of inoculated silage was slight. Inoculated silage started heating after 102 h but no had a temperature rise of more than 2 °C above the ambient temperature during 10 days exposure to air. The control silage started heating after 54 h and rich temperature more than 2 °C above the ambient temperature after 108 h. There were no significant differences in silages DM intake and in yield of energy corrected milk (ECM), but positive trends were detected for treatment effect on these parameters (Table 2). The greater (P<0.05) efficiency (NE_L 1 kg^{-1} ECM) observed when cows were fed the inoculated silage. The somatic cell count of the milk from cows fed inoculated silage was lower (P<0.05) than that of the control treatment and indicated improved hygienic quality of milk relative to inoculation. It is well known that the somatic cell count is a polyfactorial parameter (Pennington, 2011).

Table 1. Effect of inoculant on the fermentation characteristics of ensiled grass-legume.

Parameters	Treatments		Standard error	*P*-value[1]
	Control (C)	Inoculated (T)		
Dry matter (DM), g kg^{-1}	315.4	319.2	1.072	0.079
Crude protein, g kg^{-1} DM	149.4	159.0	1.732	*
Crude ash, g kg^{-1} DM	70.7	71.2	0.997	0.826
Net energy lactation, MJ kg^{-1} DM	6,42	6.50	0.019	*
DM losses, g kg^{-1} DM	106.2	88.3	3.565	**
pH	4.38	4.25	0.023	*
Lactic acid, g kg^{-1} DM	36.74	44.15	1.419	**
Acetic acid, g kg^{-1} DM	28.23	32.17	1.021	0.051
Butyric acid, g kg^{-1} DM	2.15	0.23	0.362	**
Ethanol, g kg^{-1} DM	7.87	7.06	0.217	0.059
Ammonia N, g kg^{-1} total N	57.5	46.0	1.746	**
Aerobic stability, h	108	>240	0.964	**

[1] * and ** denote statistical significance at level 0.05 and 0.01, respectively.

Table 2. Responses of dairy cows fed inoculated or untreated silage.

Parameters[1]	Silage		Standard error	P-value[2]
	Control (C)	Inoculated (T)		
Silage DM intake, kg d^{-1}	10.7	11.4	0.288	0.225
Total DM intake, kg d^{-1}	14.7	15.4	0.11	0.988
Total NE_L intake, MJ kg d^{-1}	103.0	108.5	2.792	0.341
Milk, kg d^{-1}	16.6	17.7	0.467	0.254
ECM milk, kg d^{-1}	17.4	18.8	0.531	0.183
Milk fat, kg d^{-1}	0.72	0.78	0.024	0125
Milk protein, kg d^{-1}	0.56	0.60	0.014	0.125
NE_L /1kg ECM	5.93	5.77	0.024	**
Somatic cell count, ×1000	222.3	125.1	24.144	*

[1] DM = dry matter; ECM = energy corrected milk; NE_L = net energy lactation.

[2] * and ** denote statistical significance at level 0.05 and 0.01, respectively.

Conclusions

Applying selected lactic acid bacteria prior to ensiling improved fermentation and aerobic stability of the legume-grass silage. Inoculation had small effect on intake of silage or milk yield but utilization of feed energy and the milk hygienic quality was better with inoculated silage.

Acknowledgements

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Diet composition of grazing dairy cows with and without concentrate supplementation using different markers

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Abstract

The diet selection of dairy cows on multi-species pastures was investigated in a crossover study with 12 Swiss Holstein cows and 12 Holstein cows of New Zealand origin. All cows grazed full time and each cow was supplemented either with 0 or 6 kg concentrate per day. Both experimental periods consisted of a 21-day adaptation and a 7-day measurement period. Three marker groups, alkanes, long-chain fatty acids, and long-chain alcohols (LCOH), were analysed in feed and faeces samples to estimate diet composition based on non-negative least squares. In total seven feed groups were formed consisting of *Lolium perenne*, *Dactylis glomerata*, *Trifolium repens, Taraxacum officinale*, other grass species, other forbs and concentrate. The best discrimination of the feed was achieved with LCOH, where 91% were correctly allocated, and the poorest with all three marker groups together (12%). The average diet estimation which came nearest to the botanical composition was achieved with alkanes and LCOH including a correction for faecal marker recovery. Results using this combination indicated no differences between cow strains but an effect of concentrate supplementation on herbage selection. The aforementioned marker combination seems to discriminate satisfactorily plant species selection of dairy cows, but further validation is needed.

Keywords: diet selection, multi-species pasture, cow types, supplementation

Introduction

In temperate zones on multi-species pastures, cows have a choice about the amount and the plants they eat. Unfortunately, factors affecting feed selection of grazing dairy cows and their interactions are largely unknown (Villalba *et al.*, 2015). In contrast, substitution of herbage and reduction of grazing time due to concentrate supplementation is well documented (McCarthy *et al.*, 2007). Many studies have investigated the suitability of different cow strains or breeds for pasture-based feeding systems (McCarthy *et al.*, 2007; Piccand *et al.*, 2013), but again information about diet selection of different cow strains on pasture are rare. New Zealand Holstein cows, especially bred for pasture-based feeding systems, may have a higher feeding drive (McCarthy *et al.*, 2007), but is diet selection also affected? Plant wax markers, such as alkanes, long-chain fatty acids (LCFA), and long-chain alcohols (LCOH) are used for diet composition estimation of grazing ruminants (Dove and Mayes, 2005). Frequently, this technique is used in environments with quite diverse plant populations (rangeland species, shrubs) and is not commonly used for multi-species pastures for dairy cows in temperate climates. Therefore, the aim of the study was to test whether this approach to estimate plant species selection of dairy cows is applicable under grazing conditions in multi-species pastures in temperate climate. A further question is, which marker group or marker group combination, with or without a faecal recovery correction, delivers the most accurate diet estimation. Finally, differences between two cow strains and the impact of concentrate supplementation on plant species selection were investigated.

Materials and methods

The experiment, carried out on the organic farm 'Ferme Ecole de Sorens' (Fribourg, Switzerland) between May and July 2013, was a crossover study with two adaptation periods of 21 days and two data collection periods of 7 days. Twelve matched pairs of Swiss Holstein cows (HCH) and Holstein cows of New

Zealand origin were formed according to the number of lactation, days in milk and, for primiparous cows only, age. All 24 dairy cows were managed as a single group and grazed full-time. Paddocks were rotationally grazed for 1 to 5 days. The overall average pre-grazing and post-grazing sward surface height, measured with the C-Dax pasture meter, were 89 (standard deviation (SD) 28) mm and 58 (SD 7) mm, respectively. The pastures were long-established (at least 12 years). In addition to pasture, each cow received per day either 0 or 6 kg (as fed basis) of concentrate (8 MJ net energy for lactation and 128 g crude protein kg^{-1} DM (dry matter), labelled with octacosane) in two equal meals. Individual feed intake was estimated with the *n*-alkane technique with dosed dotriacontane and tritriacontane. Grazing and rumination behaviour were recorded automatically using a jaw movement recorder (Datalogger MSR 145, MSR Electronics GmbH, Hengart, Switzerland). During the data collection periods, 2 herbage strips (each 8×1 m) per paddock were cut before the cows entered in the paddock for the determination of the botanical composition and to collect plant species or groups of plants for analysis. Dominant plant species were *Lolium perenne* (LP), *Dactylis glomerata* (DG), *Trifolium repens* (TR), and *Taraxacum officiale* (TO). Other grass species were merged as 'other grass species' (OG) representing *Phleum pretense, Poa pratensis, Poa annua, Festuca pratensis, Agrostis* spp. and *Holcus lanatus*. Furthermore, forbs were merged as 'other forbs' (OF) representing *Plantago lanceolata*, *Ranunculus acris* and *Rumex acetosa*. The average proportions of the plant species or groups over all subsamples of all data collection weeks were 27.8% LP, 6.1% DG, 38.2% OG, 10.4% TR, 9.0% TO, and 8.5% OF in fresh matter. Diet composition of each animal was estimated using the 'EatWhat' software (Dove and Moore, 1995). Estimations were performed with alkanes, LCFA, and LCOH alone, and their combinations. For each marker the faecal recovery relative to dosed Ytterbium (rFR) was determined. All diet composition estimations were performed either without or with rFR correction. Two approaches have been used to calculate the ingested amount of each marker for rFR determination, identified respectively as rFR1 and rFR2. The first approach was based on the alkane contents of the individual plant species and the individual cow faeces and the second on the average botanical composition of all examined herbage samples. Production data were analysed using a linear mixed model.

Results and discussion

Supplemented cows produced more milk (P<0.001, 25.0 vs 22.3 kg d^{-1}). Due to substitution, supplemented cows had a lower herbage DM intake (P<0.001, 11.5 vs 15.0 kg d^{-1}) compared to non-supplemented cows, but total DM intake was higher for supplemented cows (P<0.001, 16.7 vs 15.0 kg d^{-1}). Daily grazing time (P<0.001, 466 vs 550 min d^{-1}) and grazing mastication rate (P=0.008, 72 vs 75 min^{-1}) were lower for supplemented cows, but no differences occurred between cow strains. Concentrate supplementation did not influence rumination behaviour, but HCH cows tended to ruminate less (P=0.09, 410 vs 442 min d^{-1}). The alkane contents (carbon chain length (CCL) 24 to 33; DM basis) varied between 0 and 158 mg, the LCFA (even CCL 22 to 34) between 0 and 1,367 mg and the LCOH (even CCL 20 to 30) between 0 and 6,519 mg across all plant species or plant groups and the concentrate. The most accurate discrimination of diet components was achieved with LCOH, where, based on a jack-knifed classification matrix, 96% of the plant species or groups and concentrate were correctly allocated. The LCOH were followed by alkanes, LCFA and the combination of LCFA and LCOH, where 81% of the plant species or groups and concentrate were correctly identified. The poorest discrimination with 12% correct allocation was achieved with the combination of all three marker groups. If the results of the combination of all three marker groups are omitted, the correct allocation of plant species or groups and concentrate ranged between 71% (OG) and 100% (concentrate), except for LP with 54%. Frequently, LP was mixed up with OG. The calculated rFR led to realistic values for alkanes and LCOH, but not for LCFA with CCL greater than 26. Only minor differences were found between values of rFR1 and rFR2. Diet composition was estimated with a total of 21 combinations, consisting of seven marker combinations either without or with rFR correction. The results were compared to the overall botanical composition, as pastures were evenly grazed without conspicuous refusals, and ranked by the Aitchison distance to find the nearest combination of diet choice estimation. The combination of alkanes, LCOH and rFR1 achieved the nearest estimation followed by the same marker combination with the rFR2 correction. With the first mentioned combination, differences in diet choices between the two cow strains and the concentrate supplementation treatments were investigated. No difference (P=0.49, R package 'compositions') was found between the two cow strains. However, concentrate supplementation seemed to influence the diet choice on pasture (P=0.02). Non-supplemented cows had less TR (P<0.05, robust linear mixed model from R) in their diet compared to supplemented cows. This raises the question of whether this change is caused by a specific appetite for protein due to the energy-rich supplementation or by the need to stabilise rumen fermentation (reward related behaviour) or occurs at random. An additional explanation may be that cows, known for their partial clover preference (Rutter, 2010), ate first all the clover and since the non-supplemented cows grazed longer per day, they ate more of the remaining sward, which was low in clover. The knowledge of diet selection and foraging behaviour gained with this approach, may allow optimization of the offer (herbage) to the demands of the cows, which is expected to improve animal health, welfare and efficiency.

Conclusions

The method using alkanes, LCOH and rFR to investigate diet selection of grazing cows seems to discriminate well between the individual plant species or plant groups. Further validation is needed before this method can be recommended.

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Impact of N fertilisation and legume sowing density on cereals – peas intercropping performances

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Abstract

To control weeds, lodging, parasites development, grain legumes are usually cropped with cereals to increase protein autonomy in organic farming systems. Now, the management of such intercrops needs several choices in terms of plant densities or nitrogen fertilization. These choices will depend of the main aim pursue by the farmer, e.g. (1) to produce legumes and improve farming systems protein autonomy, (2) to improve the quality, the N content of the cereal, etc. Trials mobilised in this contribution, performed during two years, in organic farming systems, explored, for winter intercrops, the impact of pea (*Pisum sativum* L.) type (fodder or protein peas) and sowing density (3 levels) and of fertilisation schemes (3 levels) on intercrop yield, on protein content, on cereal protein content and on legume/cereal proportions in the harvested mixture. Based on these results, the occurrence of N fertilization or of a high level of N availability in the soil reduce the competitive ability of the legume species in the association without increasing or even securing the yield. At the opposite, the increase of legume sowing density in the mixture has a significant impact not on the yield but on the legume proportion in the mixture harvested and on the protein content of the mixture; points of interest to improve livestock farming systems protein autonomy; and of the companion cereal, and so its quality, allowing its valorization in value chain with higher added value.

Keywords: ecological intensification, cereal-grain legume intercrop, crop competition, protein concentration, yield components

Introduction

Diversification of cropping systems by increasing the number of cultivated species and including a larger proportion of legumes was proposed as a global response to the challenges of future agriculture (Malézieux *et al.,* 2009). Indeed, instead of using synthetic nitrogen fertilizers to increase farmland productivity, innovative farming systems N fertility should be based on symbiotic N_2 fixation by legumes (Bedoussac *et al.,* 2015). Therefore, legumes are of a particular interest in organic cropping systems; that could be regarded as one prototype to enhance the sustainability of present agriculture; to maintain production yield and quality (protein content) (Bedoussac and Juste, 2010). Nevertheless, grain legumes could be sensitive to water stress, to lodging (pea, …), to weed competition and/or to pest and pathogens, especially when cropped in pure stand. To face these limitations, cereal/legume intercrops, with species sown and harvested together, is an interesting option (Bedoussac *et al.,* 2015).

Now, the management of such intercrops needs several choices in terms of varieties, plant densities and patterns, and nitrogen fertilization. These choices will depend of the main aim pursue by the farmer: (1) to improve the quality, the N content of the cereal, (2) to produce legumes and improve farming systems and European plant protein autonomy; in such a case the main role for the cereal is to reduce weed pressure and the spread of diseases and pests by the physical barrier effect and, in the case of peas, to provide support to avoid lodging; (3) to sustain soil fertility, needing to define intercrops subsequent effects and minimum time of return in the rotation (Bedoussac *et al.,* 2015).

Trials mobilised in this contribution, in organic farming systems, explored, for winter intercrops, the impact (1) of pea (*Pisum sativum* L.) sowing density and (2) of fertilisation schemes on intercrop yield, on protein yield, on cereal protein content and on legume/cereal proportions in the harvested mixture.

Materials and methods

Trials were conducted in Belgium,, in 2014 and 2015, under an oceanic-mid mountain climate (49°60'N – 5°30'E; 500 masl; 1,100 mm mean annual precipitation; 7.2 °C mean annual temperature). Trials were performed in split-plot designs, with pea type (fodder pea Arkta and protein pea James (2014) and Enduro (2015)) as main parcel. Legume sowing densities (low/intermediate/high: 60, 80 and 100 seeds m^{-2} and 15(10), 20(15) and 25(20) seeds m^{-2}, respectively for protein pea and fodder pea, in 2014(2015) for this last one) × N treatments (3 levels: 0, 40 and 80 kg N ha^{-1}) being randomly distributed within these main parcels. There were 4 locks per trial. Triticale (cultivar *Sequenz* and *Borodine*, respectively in 2014 and 2015) was sowed at 350 seeds m^{-2} and 210 seeds m^{-2} respectively in association with fodder and protein peas. They were set up in fields with spelt as previous crop. Elementary parcels measured 18 m^2. NPK fertilizer (Orgamine: 7-5-10) was applied the 21 of Marsh and 22 of April, respectively in 2014 and 2015, in parallel to a light harrowing to control weeds. Harvests were done the 21 and 11 of Augustus, respectively in 2014 and 2015. A sample of about 2 kg per parcel was collected to quantify, by near infrared spectrometry, dry matter, ash, protein, starch, and cellulose contents. The part of the sample not used for the analysis was sorted to quantify pea and cereal fractions that were analysed separately to define the impact of intercropping on pea and cereal fractions qualities. Statistical analysis (took into account pea type, the sowing rate × N supply and the block factors) were performed using SAS software.

Results

Yields recorded in 2014 (1,929 and 3,272 kg ha^{-1}), were lower than 2015 yields (6,652 and 6,796 kg ha^{-1}), respectively for fodder and protein pea associations. Across both years, these yields were not impacted neither by pea sowing density nor by N treatment (P>0.19). Nevertheless, associations with fodder pea led to significantly lower yield (P<0.04), especially in 2014 (-41.0%) where we observed a trend (P=0.08) of yield decrease in parallel to legume sowing density increase. In 2015 this difference was only of 2.2%. Due to the huge contrast existing among both years, the following parameters were analysed year per year.

In 2014, the average pea content was of 70%, with an impact of pea type (P<0.01) (74 and 65% respectively for fodder and protein peas). In 2015, the average pea content was of 41%, with a reverse impact of pea type (30 and 51% respectively for fodder and protein peas). At the opposite to yields, harvested association compositions, in terms of pea/cereal proportions, mixture protein content and cereal protein content, were significantly impacted by sowing densities and/or N fertilisation (Table 1). In average, an increase of legume sowing density, by more than 60%, led to a parallel increase of 10% [5 to 17%] of the pea content and of 1.6% [1.1-2.3%] of the protein content in the harvested mixture. Nevertheless, main part of the increase (75% for pea content in the harvested mixture and 66% for protein content) occurred when shifting from the low to the intermediate densities. An increase of pea sowing densities also led to an average and significant increase of triticale protein content of 0.75% [0.5 to 1.2%]. In parallel, N supplies decreased pea proportion and protein content in the harvested mixture, in a significant way in 2014, without significant impact on triticale protein content.

Discussion and conclusions

Two non-exclusive hypotheses can be drawn to explain between years differences in terms of pea content: (1) the yellow rust pressure exerted on triticale in 2014 and/or (2) a poorer soil in 2014 site. Indeed, a difference of between years sites fertility could partly explain a high pea proportion in the harvested mixture, already at low pea sowing density in 2014, and the occurrence of pea proportion response to N supplies in 2014 and not in 2015. The occurrence of a parcel of triticale without pea would have allowed

Table 1. Impact of N fertilisation and legume sowing density on cereals-grain legumes mixtures characteristics. Pea sowing density × N supply interaction was significant (P<0.02), with fodder pea, for protein content and pea content, respectively in 2014 and 2015.

	Pea content (%DM basis)[1]				Protein content (%)				Cereal protein content (%)			
	Fodder		Protein		Fodder		Protein		Fodder		Protein	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Pea sowing density												
Low	71	24b	61b	42b	23.0b	14.9c	20.4	17.6b	10.7b	12.1b	9.4b	12.5b
Inter	76	31a	67a	52a	24.0a	15.9b	21.2	19.0a	10.9b	12.1b	9.7ab	12.8ab
High	76	36a	68a	59a	24.2a	16.9a	21.5	19.9a	11.9a	12.7a	10.1a	13.0a
N supplies (kg ha^{-1})												
0	82a	33	71a	54	25.6a	16.3	22.8c	19.1	10.8	12.2	9.7	12.6
40	72b	29	67b	52	23.2b	15.7	21.2b	19.0	11.1	12.3	9.7	12.7
80	69b	29	57c	48	22.4c	15.8	19.2a	18.5	11.6	12.4	9.8	12.9

[1] DM = dry matter.

[2] Colums showing different letter are significantly different.

to validate these hypotheses. Based on these results and in line with literature (e.g. Bedoussac *et al.,* 2015), the occurrence of N fertilization reduce the competitive ability of the legume species in the association without increasing or even securing the yield: whatever the level, N fertilization had a null efficiency in terms of MS and N yield (result not shown).

The increase of legume sowing density in the mixture has a significant impact not on the yield but on the legume proportion in the mixture harvested. The positive effect of an increase in legume sowing density on (1) protein content in the harvested mixture is of interest to improve the livestock farming systems autonomy and (2) cereal protein content is of value for cereal species (wheat, spelt, ...) that can be used in supply chain with a good added value (bread production, ...).

In conclusions, these results do not support the use of organic N fertilization to secure intercrop yield. A modulation of legume sowing density, even if not improving yield, allows to improve (1) pea and protein proportion in the harvested crop, of interest to improve livestock farming systems protein autonomy and (2) cereal quality (protein content) for cereals valorized in value chain with a high added value.

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Rejection of grass around dung pats; influence of smell, taste or both?

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Abstract

Dung pats reduce the surface of grass growth and cows reject grass around these pats, which decreases forage area and lowers the utilization of the pasture. The question is whether the rejection is due to smell, taste or both. In four grassland plots four treatments (six dung pats per treatment) were assigned with artificial dung pats; (1) untreated (control), (2) smell absent, taste present, (3) smell present, taste absent, (4) smell absent, taste absent. Three weeks after introduction of the dung pats, 10 cows were introduced in a plot during daytime on four consecutive days (new plot each day). Grass length and grazed surface around the dung pats was measured before and after grazing. The results show that smell, and the combination of smell and taste has a negative effect on the intake of grass. These results are of interest for dung pat management on grassland.

Keywords: dairy cows, dung pats, grass surface, pasture utilization

Introduction

Fouling of pastures by cow-dung pats is an important problem at dairy farms. A cow produces around 8 till 15 dung pats per day (Castle and MacDaid, 1972; MacDiarmad and Watkin, 1972; Dohi *et al.*, 1991), each covering an area of approximately 0.05-0.07 m^2 (MacDiarmad and Watkin, 1972). Cows reject herbage around pats, for more than a month, which decreases the area on which cows forage and lowers the utilization of the pasture (Bosker *et al.,* 2002; Castle and MacDaid 1972; Dohi *et al.,* 1991). In general pasture studies focussed on the effect of smell of the dung (Bosker *et al.*, 2002; Marsh and Campling, 1970) whether or not in combination with visual appearance (MacDiarmad and Watkin, 1972). In this study we investigated whether cows reject grass around dung pats due to smell, taste or both.

Materials and methods

The experiment was carried out in the growing season of 2014 in the Western peat soil district in the Netherlands. Three days prior to the start of the experiment fresh dung was collected direct from the cows' rectum, mixed and stored at 7 °C. The cows from which the dung was collected received a fresh grass and concentrate ration, and were not treated with anthelmintics and antibiotics. The experiment was set-up as a randomized complete block design with 4 pasture plots of 6.5×6.5 m as blocks and dung treatment as plot (4 treatments, 6 replicates). Dung was weighted and poured into round pie tins in order to provide the same shape of each dung pat. The following treatments were assigned; (1) smell present, taste present (control), (2) smell absent, taste present, (3) smell present, taste absent, (4) smell absent, taste absent. The absence of smell was realised by covering dung pats during grazing. The absence of taste was realised by placing the dung pats in a PVC tube that was 20 cm inserted in the ground (no nutrient leakage). Three weeks after introduction of the dung pats, 10 dried off Holstein Friesian dairy cows were introduced in a field plot between 8 AM and 5 PM on four consecutive days (repetition; new field plot each day; around 110 kg dry matter cow^{-1} per 9 hours, *ad libitum* access to water). During grazing the behaviour (walking, resting, grazing, ruminating and displacement) of the cows was observed. Cow dung excreted during the observation period was immediately removed after the observation period. Three weeks after the observational period (round 1) the experiment was repeated in the same plots with the same cows (round 2). Grass length around the dung pats (distance of 5 cm to dung pat) was measured before and after grazing (4 quartiles per pat). From this the grazed grass length was calculated. Grazed surface around the dung pats was measured after the grazing period with project developed software. Grazed grass length and grazed surface around the dung pats were analysed by analysis of variance (ANOVA) using SPSS 18.0.

Results

The uptake of tall grass differed significantly (P=0.043) between the two rounds. In round 1 grazed grass length was significantly (P≤0.0001) different for the first observation day and no treatment effects were revealed. From the behaviour of the cows (excessive walking through the plot) it could be concluded that the cows had to get used to the experimental set-up. For this treatment effects on grazed grass length were analysed for observation day 2, 3 and 4. No differences existed in grazed grass length, therefore the data of these days were combined and analysed for treatment effects. The grazed grass length of the treatments in which smell was present was significantly (P≤0.05) lower than treatments in which both smell and taste were absent.

In round 2 the uptake of tall grass differed significantly (P≤0.000) between the second observation day in comparison to the other days. Day 2 was characterised by temperatures around 35 °C, in which the cows were not motivated to graze. For reasons of animal welfare the cows were housed indoors before 5 PM. For this reason treatment effects on grazed grass length were analysed for observation day 1, 3 and 4. No differences existed in grazed grass length, therefore the data of these days were combined and analysed for treatment effects. No treatment effects were found (P≤0.119). The grazed grass length results are presented in Figure 1.

Differences in grazed surface around the dung pats were analysed per round, as this parameter was not measured in round 1 on the first observation day. Within round 1 and round 2 no significant differences existed between the plots (P≥0.05). Therefore, for both rounds, data were combined for all plots and tested for treatment effects. No treatment effects were found in round 1 (P=0.082). In round 2 grazed surface around the dung pat of the control treatment was significantly (P≤0.000) higher than the other treatments. Results on grazed surface around dung pats are presented in Figure 2.

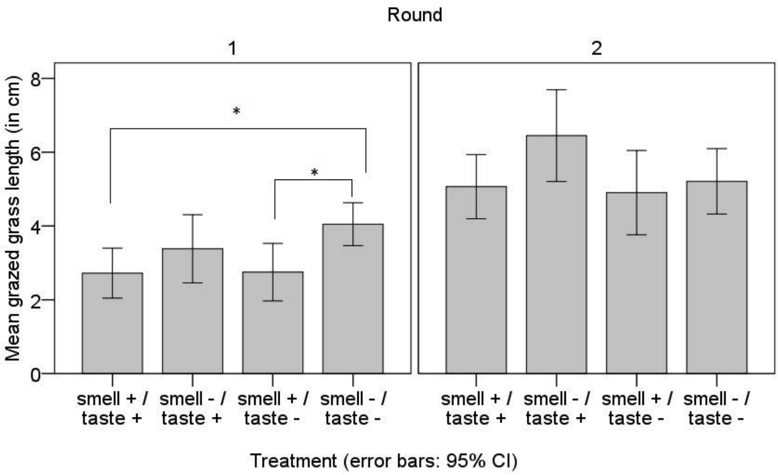

Figure 1. Mean grazed grass length of tall grass around dung pat.

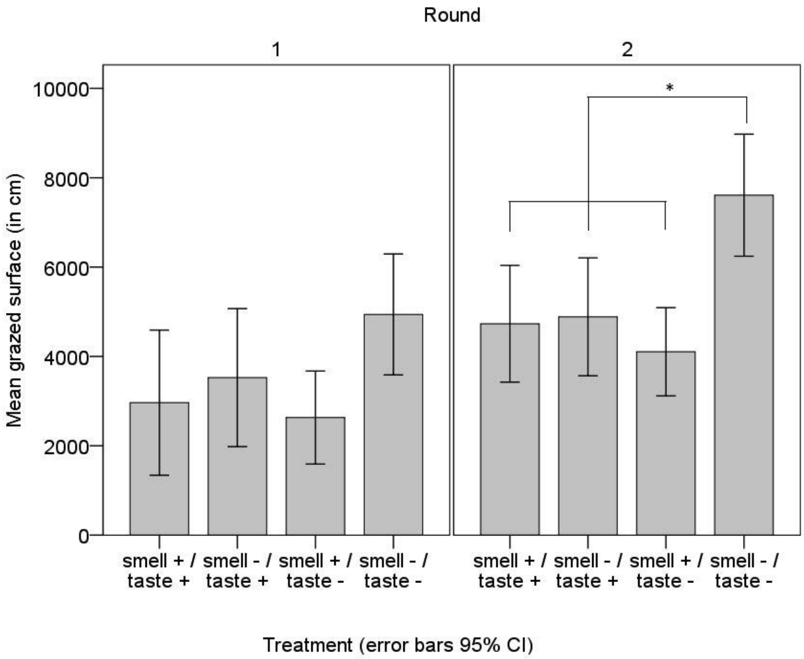

Figure 2. Mean grazed surface around dung pat.

Discussion and conclusions

The results of this study show that smell is the main factor for cows to reject tall grass around dung pats, while smell and/or taste reduces the grazed surface around dung pats. These findings are in line with findings of Marsh and Campling (1970) and MacDiarmad and Watkin (1972). The differences in uptake of tall grass could not be explained by the feeding value and mineral content of these grasses, as these were not significantly different between the treatments (results not shown). This might explain why tall grass around dung pats were not completely neglected, which is in line with findings of Bosker *et al.* (2002). Results on the effect of smell and/or taste could have been more pronounced if dung pats without smell and taste were left out, in order to stimulate the cows to make a choice between smell or taste. In conclusion it is recommended to direct pasture management at those measures which optimise rapid disappearance of the dung pat and its smell, e.g. by means of harrowing with watering (unpublished results).

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Protein efficiency in grazing dairy cows supplemented with partial mixed rations with or without legumes

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Abstract

Achieving a viable food production through a sustainable management of natural resources is the main aim on the new European Common Agricultural Policy. In this way, legumes are proposed as alternative to grasses because they contribute to get protein self-sufficiency to the farms and they require less N fertilization and water. The goal of the present experiment was to study the effects on protein efficiency of faba bean-rapeseed or Italian ryegrass silages included in dairy cows' rations. Two trials (autumn and spring) were carried out following a cross-over design with isoenergetic and isoprotein partial mixed rations complemented with herbage on pasture offered to 10 Holstein cows. Dry matter intakes were similar for both treatments. Milk fat was higher and milk protein was lower in autumn than in spring without differences between silages. Our results showed that the protein efficiency was not significantly altered by treatment (29.35%, on average). Despite this, milk urea concentration was lower for cows fed Italian ryegrass silage than faba bean-rapeseed silage (253 vs 337 mg l^{-1}). It is concluded that faba bean-rapeseed silage could be a substitute for Italian ryegrass silage, with similar protein efficiency when they are used as a supplement for grazing cows.

Keywords: N use efficiency, faba bean-rapeseed, Italian ryegrass, silages, milk

Introduction

The increased interest in sustainable livestock production systems that also are productive, are reflected in the recent reform of the Common Agricultural Policy (CAP) in Europe, that proposes new concepts involving changes in cropping of arable lands. To strengthen the environmental sustainability of animal production systems, the CAP stimulates the use of natural resources (i.e. grazing) and crop diversification ('greening') including legumes as alternative crops, among others. Legumes play an important role as nitrogen-fixing crops, contributing to reduce the protein supplements in the livestock rations and to attain protein self-sufficiency. Moreover, they are related to the environmental sustainability, because they enable to reduce the rate of N fertilization and water requirements, in comparison with grasses (Ferreyrolle, 2016). The objective of the present study was to examine the effects on protein efficiency in grazing dairy cows' supplemented with two different silages included in the rations: an Italian ryegrass (*Lolium multiflorum* Lam.) silage and an alternative intercrop silage consisted of a mixture of faba bean (*Vicia faba* L.) and rapeseed (*Brassica napus* L.).

Materials and methods

Two trials were conducted in autumn 2013 and spring 2015 at the SERIDA experimental farm (Villaviciosa, Asturias, Spain). Two adjacent plots (1.7 ha each one) were seeded with the tested winter crops (Italian ryegrass and a faba bean-rapeseed intercrop, respectively). The fertilization (chemical for Italian ryegrass vs organic for the intercrop), seed rates, sampling and analyses of both crops are described in Jiménez *et al.* (2014). Both crops were used to make silages, which were included in two different dairy cows' partial mixed rations (PMR). Ten homogeneous Holstein dairy cows (two groups with five cows in each) were used in each trial. Cows were randomly assigned to both rations following a cross-over design (two periods of 21 days each, including 14 days to dietary adaptation and 7 sampling days). Each group was fed *ad libitum* with PMR formulated according to requirements of NRC (2001), being composed mainly by Italian ryegrass silage (PMR IR) or faba bean-rapeseed intercrop silage (PMR FBR). Furthermore, cows grazed eight hours daily and ate an extra concentrate during the milking sessions. The nutritional values of each PMR, concentrate and pasture are shown in Table 1. After the adaptation period, the individual intakes and milk production were recorded daily during 7 days. Herbage intakes on pasture were estimated following the animal performance method (Macoon *et al.*, 2003). Each PMR was sampled daily and the samples were pooled to obtain a final PMR sample. Paddocks were sampled the first and the last day in each assay period to analyse the grass nutritive quality. Both PMR and grass were dried at 60 °C during 24 h for dry matter (DM), ground (0.75 mm) and analysed by near infrared spectrometry (NIRS) for organic matter and crude protein. The extra-concentrate was also milled (1 mm) and analysed by NIRS. Milk was sampled in morning and afternoon milking sessions during 3 days. Milk samples were analysed by MilkoScan FT6000 to determine the fat, protein and urea content. The protein efficiency was calculated as g milk protein N g^{-1} N intake. Intakes, milk yields, milk composition and protein efficiency were analysed for both trials by analysis of variance (ANOVA) using R statistical package (R Core Team, 2014) with silage as fixed effect. The results are shown as means of both trials.

Table 1. Chemical composition (g kg^{-1} of dry matter) and net energy of lactation (NE_L; MJ kg^{-1} of dry matter) of partial mixed rations based on Italian ryegrass (PMR IR) or faba bean-rapeseed (PMR FBR) silages, concentrate and herbage from the pastures in autumn and spring.

	Autumn				Spring			
	PMR IR	PMR FBR	Concentrate	Herbage	PMR IR	PMR FBR	Concentrate	Herbage
Dry Matter	416.0	408.1	884.9	155.2	375.5	418.1	874.5	135.1
Organic Matter	896.2	893.4	930.9	893.9	897.5	898.6	927.3	895.8
Crude Protein	127.4	133.3	194.1	160.9	159.1	158.6	171.7	219.4
NE_L	6.15	6.11	8.20	5.86	6.07	6.15	8.12	7.03

Table 2. Daily dry matter (DM) intake per cow, milk performances and protein efficiency for two rations based on Italian ryegrass (PMR IR) or faba bean-rapeseed (PMR FBR).

	PMR IR	PMR FBR	r.s.d.[1]	Significance[1]
Intake				
PMR (kg DM)	11.7	11.8	2.81	NS
Concentrate (kg DM)	2.84	2.75	0.660	NS
Herbage on pasture (kg DM)	7.05	4.58	5.931	NS
Total (kg DM)	21.6	19.1	5.46	NS
N intake (g d^{-1})	550	494	156.5	NS
Milk yield and composition				
Milk yield (kg d^{-1})	30.7	28.7	6.20	NS
Fat (g kg^{-1})	36.9	36.6	2.52	NS
Protein (g kg^{-1})	31.5	30.7	1.83	NS
Urea (mg l^{-1})	253	337	47.6	***
Protein yield (kg d^{-1})	0.96	0.88	0.166	NS
Milk protein N (g d^{-1})	151	138	26.0	NS
Protein efficiency (%)	29.1	29.6	7.92	NS

[1] r.s.d. = residual standard deviation. Significance level: NS = P>0.05; *** = P<0.001.

Results and discussion

Dry matter intakes and milk performances are presented as means for the two experiments in Table 2. Herbage, PMR and total intakes were unaffected by silage types, which was reflected in similar amounts of N intakes (522 g day^{-1} on average). Milk yields were similar in both experiments and not affected by treatment (29.7 kg d^{-1}). We did not detect any effect of treatment on milk fat or protein concentration in the trials. However, milk fat content increased 3.2 g kg^{-1} in autumn trial respective to spring trial, according to Bertocchi *et al.* (2014). Conversely, milk protein content decreased 1.7 g kg^{-1} in autumn respective to spring, which could be due to a better quality of the herbage protein in spring. Despite this, protein yield did not show differences between treatments. For this reason, secretion of milk protein N (milk true protein / 6.38; Olmos-Colmenero and Broderick, 2006) was similar for both treatments (144 g day^{-1} on average). The average of protein efficiency was 29.35%, which is consistent with the regular value of diet N retained (Edouard *et al.*, 2016). Despite this similarity in the efficiency of protein utilisation, the milk urea content showed differences between treatments. It was influenced by treatment, with higher concentrations when cows were fed PMR FBR than PMR IR (337 vs 253 mg l^{-1}, respectively, P<0.001). This fact could be due to different silages' amino acid composition or high ammonia N in faba bean silage than Italian ryegrass silage (Baizán *et al.,* 2016).

Conclusions

The protein efficiency of dairy cows feeding FBR or IR silages was quite similar, without a drop in dairy cows' performance. This fact indicates that IR silage could be substituted by FBR silage in dairy cow feeding stuffs. This strategy could reduce the impacts on the environment, achieving an important goal for the reformed CAP.

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Digestibility and degradability of seaweed protein in ruminants

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Abstract

An increasing interest for locally produced feedstuff for ruminants to decrease the import of e.g. soya is the reason for looking at seaweeds as potential protein source. Therefore, eight seaweed species (three brown, three red and two green) were sampled during spring and autumn 2014 in Bodø, Norway. Samples were washed to eliminate sand and reduce exogenous salt, and freeze-dried before analysis. Samples were analysed for chemical analysis. Rumen effective protein degradability (EPD) and total tract protein digestibility were estimated *in situ* (Dacron bag and mobile bag method, respectively). Dry matter (DM) concentrations were similar to terrestrial forages (11-28% DM). Ash concentrations were high and variable. Protein concentrations varied between 8-37% DM. Spring samples were higher in protein than autumn samples. EPD varied from 24 to 64%, and intestinal digestibility of rumen escape protein from zero to 84%. Seaweed species differ extremely in feed value, but some are interesting as energy (some red and brown) and some as protein (some red and green) feeds for ruminants. Salt concentration and ash composition might limit the proportion of seaweed of the total diet to animals.

Keywords: *in situ*, cow, crude protein, Norway

Introduction

The Norwegian meat production is highly dependent on imported soya as protein source. An increasing interest for locally produced feedstuff for ruminants is the background for the project 'Legumes and seaweed as alternative protein sources for sheep' that was launched in 2014. The main aim of the project is to identify alternative protein sources, where locally produced legumes and seaweed seem to be the most suitable, sustainable and environmental friendly sources. The potential biomass production from the sea has renewed the interest in using seaweeds as animal feed. Some seaweeds are protein rich (Stadtlander *et al.*, 2013) and therefore of interest as an alternative to soybeans. Commercial seaweed cultivation (19 million tons in 2010) mainly takes place in Asia, and only a small fraction is used for animal feed (Evans and Critchley, 2014). The published information on nutritive value of seaweeds for animals varies widely depending on the seaweed and animal species. *In vivo* data on protein degradability of seaweeds in dairy cows are scarce. The aim of this paper is to summarize *in situ* rumen and total tract digestibility of seasonal and species-specific seaweed protein in dairy cows (Tayyab *et al.*, 2016).

Materials and methods

Eight seaweed species (brown: *Alaria esculenta*, *Laminaria digitata*, *Pelvetia canaliculata*; red: *Mastocarpus stellatus*, *Palmaria palmata*, *Porphyra* sp.; green*: Acrosiphonia* sp., and *Ulva* sp.) were sampled by hand picking in spring (March) and autumn (October and November) 2014 at the coast of Bodø in northern Norway (67°19'00'' N, 14°28'60' E at low tide). *Ulva* sp. was only sampled in autumn. Seaweeds were thoroughly washed and cleaned of sand, epibionts, associated mesograzers and other vertebrates and invertebrates in baths of ambient seawater (salinity 34 mg l^{-1}) and later in fresh water. Samples were frozen at -20 °C until freeze drying. Dried samples were milled through a 1.5 mm screen with a cutter mill (Fritsch pulverisette 15) for *in situ* analysis, and a 1 mm screen for chemical analysis.

The seaweed samples (n=15) were incubated in the rumen in three dry rumen fistulated Danish Holstein cows (replicates) for *in situ* assessment of rumen effective protein degradation at 8 time intervals (0, 2, 4, 8, 16, 24, 48 and 96 h) using Dacron bags (38 µm pore size). For total tract protein digestibility, the mobile bag method was used according to Hvelplund and Weisbjerg (2000). Each seaweed sample was replicated twice in each cow. All seaweed samples were analysed for chemical composition (dry matter (DM) concentration, ash, crude protein (CP) and neutral detergent fibre (NDF) (Tayyab *et al.*, 2016). Data were analysed using PROC Mixed Model by SAS® 9.4 version (SAS Institute Inc.) with species and season as fixed effects and cow as random effect. Results are presented as average of spring and autumn samples. For details see Tayyab *et al.* (2016).

Results and discussion

Dry matter concentration ranged between 11 and 28% that is similar to many terrestrial forages. The ash concentration was generally high with large variations between species (13-48% of DM). *Porphyra* sp. contained the highest content of CP (35%) and *Pelvetia canaliculata* the lowest (8%) (P<0.0001). Consistent with terrestrial forages, protein content was higher in spring samples compared to autumn samples and the NDF content was higher in autumn samples compared to spring samples (Tayyab *et al.*, 2016).

Figure 1 shows the concentration of seaweed CP (across both seasons) that were degraded in rumen or small intestine, or passed out in faeces (indigestible). Most of the protein in forages is normally degraded in the rumen by the microorganisms and thus a minor fraction reaches the small intestines eventually to be digested and absorbed by the animal. Thus, the *in situ* techniques are used to establish the protein value of feed for the ruminants, as degradation profiles in the rumen and post rumen digestibility of rumen escaped protein. Our results show, that *Porphyra* sp. and *Acrosiphonia* sp. provided a high supply of rumen escape protein. However, *Acrosiphonia* had a higher indigestible protein fraction compared to *Porphyra* sp. and thereby *Porphyra* sp. had the highest supply of digestible protein in the small intestine (Figure 1). *Ulva* sp. had a low supply of protein to the rumen in addition to a low fraction of indigestible protein with the majority of the protein digested in the small intestine. About half of the total protein intake from

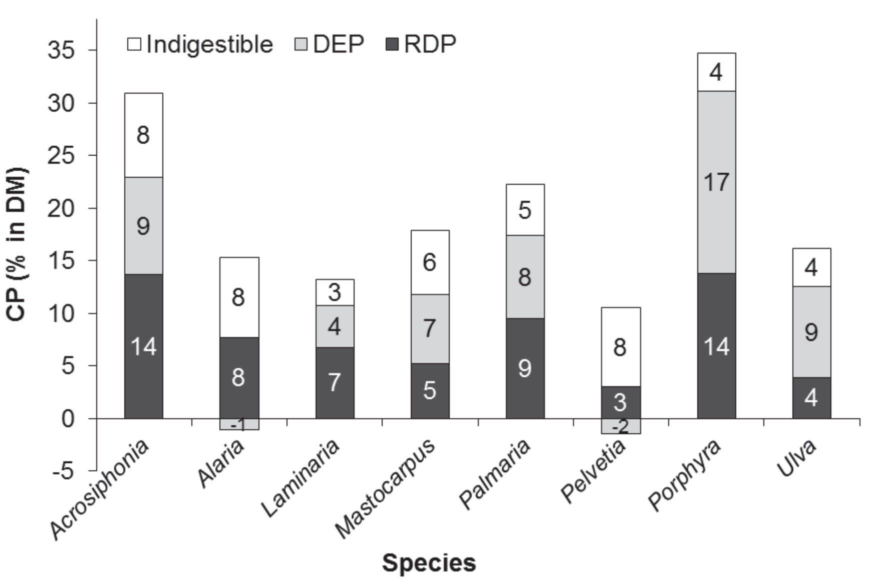

Figure 1. Crude protein (CP) as percent in degradable dry matter (DM) in eight seaweed species (herewith classified as genus; species names are in text). RDP = rumen degradable protein; DEP = digestible escape protein of the total CP intake; indigestible CP (Tayyab *et al.*, 2016).

Porphyra sp. and *Ulva* sp. was digested in the small intestine. From these results, the red seaweed species *Porphyra* sp. and the green seaweed species *Ulva* sp. and *Acrosiphonia* sp. are interesting sources of protein for ruminants based on their chemical composition and digestibility patterns. However, the protein composition, especially amino acid composition and proportion of true CP is yet to be examined for further understanding of protein values in the seaweed species. The other five seaweed species investigated were not considered of further interest as a protein source for ruminants. The next step will be to feed seaweed *in vivo* and measure, e.g. lambs growth rate.

Conclusions

In conclusion, one red seaweed species *Porphyra* sp. and two green seaweed species *Ulva* sp. and *Acrosiphonia* sp. have potential as alternative protein sources for ruminants.

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Five successful strategies for grazing in combination with automatic milking

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Abstract

About 20% of the 17,000 Dutch dairy farms use an automatic milking system. About 50% of them practise grazing while the national average is about 80%. The potential for grazing is related to the farm infrastructure (here expressed as stocking rate per ha grazing area) and to the personal preferences and experiences of the farmer. Five coherent and promising grazing strategies for farms with automatic milking have been developed. 'Maximum grazed grass' can be used for stocking rates up to 2 cows per ha grazing area; 'Plenty of grazed grass' can be used up to 3 cows; 'Half and half grazed grass' can be used up to 5 cows; 'Grazed grass as supplement' for up to 7 cows and 'Grazed grass as dessert' for up to 10 cows. For each grazing strategy data are provided on stocking rate, grass intake, hours grazing, supplementation, optimal grazing system, farm layout, daily management and on key indicators for personal preferences.

Keywords: automatic milking, dairy, grazing, grazing strategy, grazing systems

Introduction

About 20% of all 17,000 Dutch dairy farmers currently use an automatic milking system (KOM, 2016). This percentage is expected to increase to 30% in 2020 (Van den Pol-Van Dasselaar *et al.*, 2015). There is a clear relation between the type of milking system and whether or not a farmer practices grazing. The percentage of grazing at farms with an automatic milking system (milking robot) is lower compared to farms with a conventional milking system (52 vs 85%). Nationally, on average 80% of all farms currently practise grazing. The potential for grazing is further related to the stocking rate per ha grazing area (Holshof *et al.*, 2016) and the personal preferences and experiences of the farmer. It seems that farmers in the Netherlands are not keen on combining automatic milking with grazing.

In the late nineties a few research trials were done in the Netherlands with cow traffic and suitable grazing systems. However, the focus then was on technical aspects and not on embedding strategies in current farm systems. In practice, good examples of combining grazing and automatic milking are present in the Netherlands. The diversity of practices was, however, never translated in general guidelines that could easily be adopted by farmers or extension services. This study aimed to assess key indicators for successful grazing strategies for Dutch dairy farming systems and to develop general guidelines for grazing on farms with an automatic milking system. Using the experience of a diverse group of farmers that have been combining automatic milking and grazing successfully and the experience of farmers' advisory service providers, common practices were translated into a set of guidelines for a variety of farm types.

Materials and methods

This study was part of the wider program 'Robot and grazing', in which in total 500 dairy farmers and a whole range of service providers participated. For the purpose of this research, two target groups can be discerned:

- five groups, based on region or farm size, of in total 50 dairy farmers that had been combining automatic milking and grazing for at least two years; each group was led by a group facilitator with expertise on grass and grazing;
- an expert panel of farmer's advisory service providers.

The farmer-groups met during two growing seasons. They provided numerous and diverse practical experiences as input for successful grazing strategies. These experiences were used in the project by means of a specific participatory approach in which individual experiences were transformed to general explicit knowledge. First three farmer meetings were used to diverge, i.e. to obtain as much experiences as possible. Then, two farmer meetings were used to converge, i.e. to arrive at the essence of successful grazing strategies. The farmer meetings alternated with meetings of advisory services (e.g. feed industry, robot industry) which provided further valuable input to the grazing strategies.

Results and discussion

The project developed general guidelines for grazing in combination with automatic milking that can easily be adopted by farmers or extension services:

1. Five grazing strategies based on stocking density will lead to a feasible strategy for the majority of Dutch farms and farm conditions.
2. There are four key indicators of personal preferences of farmers with respect to grazing strategies; they reflect important items for a farmer.
3. Each grazing strategy can be defined using parameters of intake, grazing system and the key indicators.

The five grazing strategies are presented in Table 1. When stocking rates increase, both the number of hours grazing and the grass intake decrease and supplementation increases. For each grazing strategy, guidelines were produced with respect to grassland management and farm layout. Furthermore, examples of farm layout and daily schedules were given (not shown). The four key indicators to personal preferences, that make distinctive strategies, were:

- grass utilisation: a high score for grass utilisation indicates good opportunities for a high net grass yield, low feeding costs and low residuals;
- ease of labour: a high score for ease of labour indicates that less labour is required; the difference between high and low scores is half an hour per day;
- farmers' know-how: a high score for know-how indicates that the farmer needs more grazing skills;
- milk production: a high score for milk production indicates that milk production may be somewhat higher, but certainly more stable throughout the season.

The strategies are meant as good practice examples. They should inspire, but are not meant as a blueprint for every farm. Personal preferences are important in choosing the right strategy for a farm.

Conclusions

In this project coherent and promising grazing strategies for farms with automatic milking have been developed for different stocking rates. 'Maximum grazed grass' can be used for stocking densities up to 2 cows per ha grazing area; 'Plenty of grazed grass' can be used up to 3 cows per ha grazing area; 'Half and half grazed grass' can be used up to 5 cows per ha grazing area; 'Grazed grass as supplement' for up to 7 cows per ha grazing area and 'Grazed grass as dessert' for up to 10 cows per ha grazing area.

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Table 1. Strategies for grazing in combination with automatic milking.

	Strategies for grazing				
	1 Maximum grazed grass	2 Plenty of grazed grass	3 Half/half grazed grass	4 Grazed grass as supplement	5 Grazed grass as dessert
Stocking rate, ha^{-1}	<2	<3	<5	<7	<10
Grass intake, kg DM cow^{-1} d^{-1}	13	10	6	4	2
Hours grazing, d^{-1}	18	15	9	6	3
Roughage supplementation, kg DM cow^{-1} d^{-1}	2	5	9	11	13
Optimal grazing system[1]	Strip	Strip	Rotational set stocking	Rotational set stocking	Set stocking
Grazing gate	Yes	Yes	Yes	Yes	No
Grass (utilisation)[2]	*****	****	***	**	*
Ease of labour	*	**	****	****	*****
Farmers' know-how	*****	*****	***	***	*
Milk production	***	***(*)	****	****(*)	*****

[1] Strip = strip grazing, which is a typical rotational grazing system (rotating two or three times a day) with back fencing. Set stocking is a grazing system where the cow can graze the daily (re)growth at a platform for three till six weeks at an average grass length of 8-10 cm. Rotational set stocking is essentially identical to set stocking, but on the platform paddocks are split up and cows rotate every day within these split paddocks.

[2] Each key indicator is valued with a score from * to *****.

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Fatty acids routed from fresh grass to milk influence $\delta^{13}C$ in milk

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Abstract

Dairy production systems vary widely in their feeding and livestock-keeping regimens. Both are well-known to affect milk quality and they are important topics for consumer perceptions. Here we examine whether carbon isotope composition ($\delta^{13}C$) in milk can be sufficiently predicted from feeding information, so that measurement of $\delta^{13}C$ in milk could serve as authenticity proof. We obtained 671 milk samples from 40 farms distributed over Central Europe to measure $\delta^{13}C$ and fatty acid composition. Feeding protocols by the farmers in combination with a model based on $\delta^{13}C$ feed values from the literature were used to predict $\delta^{13}C$ in feed and subsequently in milk. The model considered dietary contributions of C_3 and C_4 plants, contribution of concentrates, altitude of forage production, seasonal variation in $^{12/13}CO_2$, Suess's effect, and diet-milk fractionation. Predicted and measured $\delta^{13}C$ in milk correlated closely (r^2=0.93). Analysing milk for $\delta^{13}C$ allowed validation of a reported C_4 component with an uncertainty of <8% in 95% of all cases. This included the uncertainties of the method (measurement and prediction) and of the feeding information. However, the mismatch was not random but varied seasonally and correlated with the seasonal variation in long-chain fatty acids originating from fresh grass. This indicated routing of long-chain fatty acids from fresh grass to milk. In conclusion, $\delta^{13}C$ in milk can be predicted in the authentication case but larger errors occur with high proportions of fresh-grass feeding, due to routing of long-chained fatty acids from fresh grass to milk.

Keywords: authentication, isotopes, unsaturated long-chain fatty acids, maize, grass

Introduction

Isotope analysis could be valuable for food authentication, proof of origin or proof of production system. In experiments showing that the isotopic composition of feed is forwarded to the product, both the feed and product are usually measured. This does not comply with the situation typical for authentication in which the isotope composition of the feed is unknown and has to be estimated from the information of the reported feeding regimen or the livestock-keeping regimen. However, for milk there are no procedures in literature on how to make such a prediction or which parameters to include. We set up such a prediction model to examine the hypothesis that the designated feeding information can be verified by analysing $\delta^{13}C$ in milk. We then quantified the prediction uncertainty that results even with correct feeding information. Further, we will analyse whether routing of fatty acids derived from fresh grass contributes significantly to the mismatch between expected and measured $\delta^{13}C$, because it is known that long-chained unsaturated fatty acids, which differ strongly in $\delta^{13}C$ (Richter *et al.,* 2012) are routed (Dewhurst *et al.,* 2006). An extended version of this work can be found elsewhere (Auerswald *et al.,* 2015).

Materials and methods

Sampling comprised 40 farms in Austria and Germany with detailed analysis of the feeding and livestock keeping regimes. Feeding protocols were obtained by interviews. In total 671 milk samples were collected year round and analysed for $\delta^{13}C$ and fatty-acids. Prediction of $\delta^{13}C$ in milk distinguished between roughage and concentrates derived from either C_3 or C_4 plants, thus:

$$\delta^{13}C_{feed} = m_a \cdot \delta^{13}C_a + m_b \cdot \delta^{13}C_b + m_c \cdot \delta^{13}C_c + m_d \cdot \delta^{13}C_d$$

where m denotes the fraction of total dry feed mass and the indices a to d refer to the four components C_3 or C_4 roughage and C_3 or C_4 concentrates. The basic $\delta^{13}C$ values of the four components (-29.5‰, -12.7‰, -26.8‰, -12.1‰) have to be adjusted for the Suess effect (which is the change in atmospheric $\delta^{13}C_{CO2}$ mainly due to fossil fuel burning since 2003 when the basic $\delta^{13}C$ values for the feed components were determined); C_3 roughage has additionally to be adjusted to the mean altitude of the individual farm and fresh C_3 roughage for the seasonal variation of $\delta^{13}C$ in atmospheric CO_2 (Table 1).

Results and discussion

Measured $\delta^{13}C$ of milk correlated closely with the independently predicted $\delta^{13}C$ of milk (r^2=0.932, n=671, P<0.001; Figure 1A) with a root of the mean square error (RMSE) of 0.75‰. The RMSE comprised less than one tenth of the entire range of feed $\delta^{13}C$. When restricting the data set to farms feeding no maize and less than 15% concentrates (12 farms; 397 milk samples), for which a mismatch between prediction and measurement could not result from errors in the reported feed ration, the percentage of fatty acids with a chain length of 18 and longer (long-chain fatty acids – LCFA; mainly unsaturated LCFA from grass) correlated with the unexplained variation of $\delta^{13}C$ in milk (r^2=0.225, n=397, P<0.001). LCFA have a much more negative $\delta^{13}C$ than shorter fatty acids (Richter *et al.,* 2012)

Table 1. Prediction model based on published $\delta^{13}C$ for specific feed components. Total feed composition is given from the mass fractions of the four components (a: C_3 roughage; c: C_3 concentrate; b: C_4 roughage; d: C_4 concentrate) and their isotopic composition. For C_3 roughage (fresh or conserved), an altitude effect was included. Parameter a denotes altitude above sea level in m. The Suess effect since 2003 has to be added to the $\delta^{13}C$ of all components. Parameter t denotes the year of interest. The $\delta^{13}C$ of fresh grass has to be adjusted for the seasonal variation of $\delta^{13}C$ in CO_2. t_{DoY} is day of the year (unit: d). For references see Auerswald *et al.* (2015).

Model component	$\delta^{13}C$ (‰)
C_3 roughage	$\delta^{13}C_a = -29.5 + 1.1 \times a/1000$
C_3 concentrate	$\delta^{13}C_c = -26.8$
C_4 roughage	$\delta^{13}C_b = -12.7$
C_4 concentrate	$\delta^{13}C_d = -12.1$
Suess effect[1]	$\delta^{13}C_a = \delta^{13}C_a - 0.02644 \times t + 52.95$
Seasonal $\delta^{13}C_{CO2}$ variation	$\delta^{13}C_a = \delta^{13}C_a\ 0.51 \times \sin((t_{DoY} - 121)/365 \times 2\pi) - 0.35$
Diet-milk fractionation	$\delta^{13}C_{milk} = (0.4 - \delta^{13}C_{feed})/(\delta^{13}C_{feed} - 1)$

[1] Due to the irregular change in $\delta^{13}C_{CO2}$ that impedes any prediction, the difference in $\delta^{13}C_{CO2}$ between the year of interest and the reference year (2003) should better be obtained from published measurements at: http://www.esrl.noaa.gov/gmd/dv/data/.

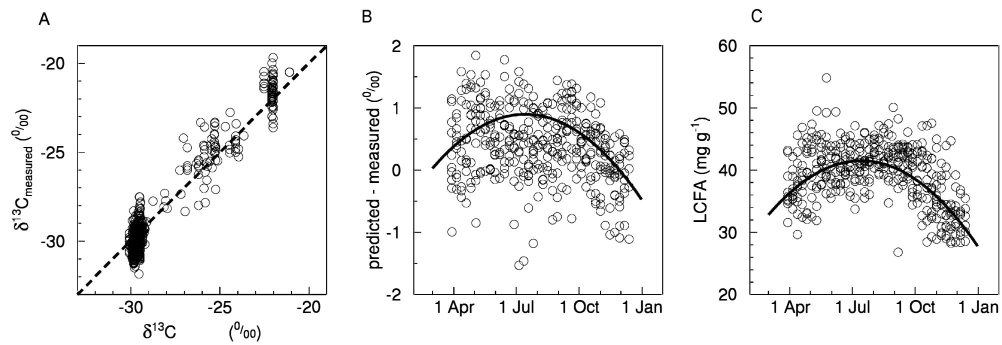

Figure 1. (A) Predicted and measured $\delta^{13}C$ in bulk milk (line denotes unity). (B) Seasonal change of predicted minus measured $\delta^{13}C$ of milk (only farms without maize feeding). The line is a parabolic regression (r^2=0.101, n=397, P<0.001). (C) Seasonal change in the proportion of fatty acids with a chain length of 18 and longer, LCFA, in total fat. The line is a parabolic regression (r^2=0.360, n=397, P<0.001).

and are not synthesized by the mammalian milk gland (Dewhurst *et al.,* 2006). The concentration of LCFA showed a distinct seasonality with a maximum in July and minima in March and December (Figure 1C). This seasonality in LCFA concentration paralleled the seasonal pattern of the unexplained variation of $\delta^{13}C$ (Figure 1B). This similar seasonal behaviour of LCFA and the unexplained variation of $\delta^{13}C$ were even more obvious when individual farms with exceptionally low contributions of concentrates were analysed separately. The regression equations between the percentage of LCFA and the residuals indicated that the $\delta^{13}C$ of LCFA should be more depleted than the average carbon in milk (by 6 ‰ to 8 ‰), which agrees with data from literature (e.g. Richter *et al.,* 2012).

In summary, a prediction model for authentication purposes was set up and proved to be suitable. Deviations between predicted and measured $\delta^{13}C$ in milk larger than 1.4‰ indicate that the error in the feeding information is larger than the uncertainty of this information on experimental farms. With large proportions of fresh grass in the diet, the routing of LCFA from grass to milk causes additional uncertainty in the prediction.

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Maize yield and composition affected by rate and timing of nitrogen fertiliser and mulch type

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Abstract

In Ireland the current nitrogen (N) input strategy for forage maize (*Zea mays* L.) is to apply and incorporate all fertiliser both organic and inorganic into the seedbed immediately before sowing. This, however, may result in losses from the cropping system by leaching, immobilisation, denitrification and clay fixation. The application of N in a split application, 50% at sowing and 50% at growth stage V6-V8 (between six to eight leaves with visible collars), may reduce the opportunity for loss and increase plant uptake and improve nitrogen use efficiency. A split-plot design was used to investigate the effects of degradable plastic mulch (Samco Green and Samco Yellow), inorganic N rate (0, 50, 100 and 150 kg N ha^{-1}) and inorganic N application timing (100% at sowing or 50% at sowing and 50% at V6-V8) on forage maize yield and chemical composition. Samco Green mulch treatment resulted in greater whole-crop (WC) yield (P<0.05), and crop nitrogen uptake (P<0.05) compared to the Samco Yellow mulch. The split application of N 50% at sowing and 50% at V6-V8 resulted in no significant increase in either WC or grain yield in comparison to 100% N applied at sowing. The present study indicates that application of N at growth stage V6-V8 did not benefit crop yield compared to the entire application at sowing.

Keywords: forage maize, plastic mulch, nitrogen

Introduction

Inorganic N fertiliser is one of the highest input costs in the production of forage maize but it is also one of the major drivers of crop yield and composition. Sheaffer *et al.* (2006) have shown a positive response in both whole-crop (WC) and grain yields to increasing N fertiliser application rates from 0 to 200 kg N ha^{-1}. There is a need to investigate different N input strategies to maize to avoid risk of losses, through leaching, immobilisation, denitrification and clay fixation (Scharf *et al.*, 2002). This is of considerable importance in Ireland and other areas of Europe where rainfall patterns can cause leaching of nitrate. Therefore, there could be merit in synchronising N supply with the crop's requirement for N.

The maize plant typically increases its demand for N during the middle of vegetative growth (Binder *et al.*, 2000), thus an application of N at growth stage V6-V8 (between six to eight leaves with visible collars) may be a possible strategy to improve N use efficiency (NUE). Islam and Garcia (2011) indicated that application of N fertiliser at V6 is the most appropriate application timing, however Dordas *et al.* (2008) showed no benefit to N applied in a single application compared with split application (50% at planting and 50% at V8) without mulch. Currently in Ireland 80-90% of forage maize is established under degradable plastic mulch (DAFM, 2015). Thus, the effects of a split N application programme for forage maize grown with degradable plastic mulch warrants investigation. The objectives of this study were to identify the optimum N fertiliser application rate and timing for WC and grain yields, estimated nutritive value and NUE when forage maize is grown under two different degradable plastic mulch types.

Materials and methods

The experiment was conducted at University College Dublin, Lyons Research Farm (53°18 N, 6°32 W) during 2014 and 2015. The soil type was a medium clay loam classified as a grey brown podzolic. Briefly, the soil in 2014 (Year 1) had pH 7.2 and available P and K of 16 and 53 mg/l, respectively, while the soil in 2015 (Year 2) had pH 6.4 and available P and K of 7 and 29 mg/l, respectively. Soil N index was index 3 and 2, respectively, based on the preceding crop history (Coulter and Lalor, 2008). The values for pH, P and K were maintained at levels recommended by Coulter and Lalor (2008). Maize was sown on 17 April in Year 1 and 16 April in Year 2 using the Samco 3-in-1 system (Samco 2200 plastic mulch laying machine) at a depth of 50 mm and a density of 102,200 seed ha^{-1}. The hybrid was P7905 (Pioneer, Hi-bred, Inc). The experiment had a split-plot design, with the main plot as two degradable plastic mulch types Samco Green plastic mulch (GPM) and Samco Yellow plastic mulch (YPM), differing in degradation score 3 and 5, respectively. Treatment with incremental inorganic N fertiliser rates (0, 50, 100, 150 kg N ha^{-1}) applied at two timings (100% at sowing or 50% at sowing and 50% at V6-V8) represented as the sub-plots, repeated within each of three replicate blocks. Each experimental plot was 3.5×20 m and the experimental block was enclosed by a four row buffering zone. During the final harvest in October the centre 5 m of the two central rows were hand harvested at a stubble height of approximately 15 cm above ground. All plants were weighed and samples were dried in an oven with forced-air circulation at 55 °C for 48 h for dry matter (DM) determination. Representative plants were separated into cob (rachis and kernel) and stover (remainder following cob removal) to determine grain yield. Nutritive value characteristics were analysed using near-infrared spectrometry (Van Waes *et al.*, 1996) based on an *in vitro* calibration for fresh dried forage maize (Scottish Agricultural College, Penicuik, UK). The data were analysed as a 2×(4×2) split-plot using the Mixed procedure of SAS v9.3.

Results and discussion

The GPM had a higher WC yield (P<0.05) (13.5 and 12.7 t DM ha^{-1}) and crop N uptake (P<0.05) (169.2 and 159.2 kg N ha^{-1}) compared to YPM (Table 1). The YPM resulted in a significantly higher starch concentration compared to the GPM (372 and 327 g kg^{-1} DM). The application of increasing

Table 1. The effect of nitrogen fertiliser rate and timing, and mulch type, on the whole-crop yield and chemical composition of forage maize.[1]

Plastic mulch	R	T	G yield (t DM ha^{-1})	WC yield (t DM ha^{-1})	CNU (kg N ha^{-1})	NUE (kg kg^{-1})	Starch yield (t DM ha^{-1})	ME (GJ ha^{-1})	DM (g kg^{-1})	Starch (g kg^{-1} DM)	N (g kg^{-1} DM)
Yellow	0		5.67	11.82	150	70.0	4.00	12.2	262	334	12.7
	50	Sow	6.55	13.26	165	65.0	4.17	12.2	272	303	12.6
		Split	6.58	11.56	145	56.7	3.89	12.3	262	334	12.5
	100	Sow	6.97	13.31	167	52.4	4.35	12.2	258	320	12.7
		Split	6.40	12.51	156	49.2	4.56	12.3	264	352	12.5
	150	Sow	6.39	13.43	169	44.2	4.16	12.3	259	302	12.6
		Split	7.27	12.67	162	41.7	4.46	12.2	268	342	12.8
Green	0		6.68	12.65	157	65.2	4.42	12.4	265	349	12.3
	50	Sow	6.69	13.48	171	66.1	4.26	12.2	260	307	12.7
		Split	6.98	12.87	162	63.1	4.69	12.3	263	359	12.7
	100	Sow	7.55	14.12	179	55.9	4.18	12.1	274	288	12.7
		Split	7.24	13.75	168	54.3	4.56	12.2	277	326	12.2
	150	Sow	7.39	15.09	190	49.6	4.67	12.0	279	299	12.7
		Split	7.69	12.54	159	41.3	4.04	12.2	273	321	12.7
SEM			1.140	0.859	8.94	4.79	0.62	0.093	10.9	30.5	0.30
F			NS	<0.05	<0.05	0.1	NS	NS	NS	<0.05	NS
N			NS	NS	<0.05	<0.001	NS	NS	NS	NS	NS
T			NS	<0.05	<0.01	0.1	NS	<0.05	NS	<0.01	NS

[1] R = rate; T = timing; G = grain; WC = whole crop; CNU = crop nitrogen uptake; NUE = nitrogen use efficiency; ME = metabolisable energy: Sow = 100% N applied at sowing; Split = 50% N applied at sowing + 50% applied at V6-V8; F = Film; N = Nitrogen; T = application timing; DM = dry matter; SEM = standard error of the mean.

rates of N fertiliser resulted in decreased (P<0.001) NUE. There was no effect of application of N 50% at sowing and 50% at V6-V8 on grain and starch yield compared to 100% application of N at sowing. Whole-crop yield was significantly higher when N application was 100% at sowing compared to the application of N 50% at sowing and 50% at V6-V8 irrespective of the degradable plastic mulch type used. There was an increase (P<0.01) in starch concentration resulting from the application of N 50% at sowing and 50% at V6-V8 and this occurred with both plastic mulch types. Overall the results indicate that application of N 50% at sowing and 50% at V6-V8 conferred no benefit compared to 100% application of N at sowing in terms of yield. This is possibly due to the high soil mineral N content reflecting previous cropping management, evidenced by the high WC and grain yield with 0 kg N ha^{-1}.

Conclusions

In conclusion GPM resulted in higher WC yield and CNU compared to YPM. Fertiliser N applied 50% at sowing and 50% at V6-V8 conferred no benefit in yield compared to a total N application at sowing. This study was undertaken over two years and work is ongoing.

Acknowledgements

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Effect of different N and P levels on the forage yield and some yield characteristics of *Pennisetum hybridum*

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Abstract

A pot experiment was conducted to evaluate the effect of N and P fertilization on the forage yield and some other yield components of giant king grass (*Pennisetum hybridum*). The experiment comprised 5 nitrogen (0, 50, 100, 150 and 200 kg N ha^{-1}) and 3 phosphorus levels (0, 50 and 100 kg P ha^{-1}). Some characteristics such as number of tillers, plant height, fresh and dry matter yield, crude protein and root weight were tested in the study. Results indicated that the effects of N and P treatments on all tested characteristics were significant, and the combination of 150 kg N and 100 kg P ha^{-1} had the highest forage yield and quality for giant king grass production.

Keywords: giant king grass, N and P levels, forage yield and quality

Introduction

Giant king grass (GKG) (*Pennisetum hybridum*) as a field crop is a perennial grass native to Africa, and amongst tropical grasses has been the most promising, producing dry matter yields of fodder that surpass most others in that category. In tropical and sub-tropical areas it mostly used for 'cut and carry' agronomic systems. GKG is a C_4 species with a perennial life cycle and is propagated vegetatively (Hanna *et al*, 1984). It has a profuse root system, penetrating deep into the soil, and an abundance of fibrous roots spreading into the top soil horizons. GKG has the potential to grow with fewer nutrient inputs, less water and to tolerate a variety of biotic and abiotic stresses compared with maize. The fact that it is perennial means that replanting every year is not required (Kukkonen, 2009); also it is a new introduction to Turkey. Investigating crop production in this species is important not only in Turkey, but also in the southern part of Europe. The most practical and effective method to increase dry matter (DM) production in forage crops is through the addition of N. Fertilization with N can increase DM production up to three-fold in forage crops, especially grasses, depending on the annual rainfall or irrigation practices. On the other hand, root development is considered by some authors to be more dependent upon adequate P and K than on N, but insufficient information is available regarding the nutrient levels necessary to optimize both root and shoot growth in GKG plants. This preliminary study had the aim to determine the optimum N and P level of application in GKG for obtaining the highest forage yields under controlled conditions.

Materials and methods

A pot study was conducted on the experimental area of Field Crops Department, Faculty of Agriculture, Ege University, Izmir, Turkey from June to November 2013. The physical properties of the soil used were: 80.2% sand, 18.0% silt and 1.8% clay; and its chemical properties were: pH 5.83, 0.03% salt, 2.27% OM, 0.092% total N, levels of available Ca, P, K were 1,300 μg kg^{-1}, 2.54 μg kg^{-1} and 40 μg kg^{-1}, respectively. The GKG cultivar 'Paraíso' was used. Cuttings 30 cm long were selected from the lower parts of GKG rootstock plants grown in full sunlight. Leaves were trimmed and the cuttings dipped in indole-3-butyric acid hormone (concentration 4,000 μg kg^{-1}) for 3-4 s up to a height of 10-15 cm and then in fungicide (a 1:10 mixture of Benomyl and talc powder). Cuttings were then inserted into each plastic pot filled with 16 kg soil on 1 June 2013. The experiment was carried out using a randomized complete block design with 4 replications: combinations of 5 N (0, 50, 100, 150 and 200 kg N ha^{-1}) and 3 P levels (0, 50 and 100 kg P ha^{-1}) were tested on GKG crop. Half the rate of N (as ammonium sulphate) and the full rate of P (as triple superphosphate) were applied before planting, and the rest of N was applied at the 7-10 leaf stage as

NH_4NO_3. All pots, including the control (N0P0) treatment, were fertilised using 100 kg ha^{-1} potassium sulphate before planting. Moisture was maintained by watering the plants every other day with 100 or 250 ml of tap water according to the moisture content of the soil. All other agricultural practices needed during growth were carried out when required. Plants were harvested (15 cm height) after 21 weeks (4 November 2013). Plant height was measured; the number of tillers per plant was also determined on the same day as harvesting. The fresh weight of above-ground components was obtained, followed by roots. Samples were dried in a forced-air oven at 65°C for 3 days and the dry weight measured. The tissue was ground to pass through a 2 mm sieve. The crude protein (CP) content of the sward was determined using the Kjeldahl method (N%) with a conversion factor of 6.25. All data were statistically analyzed using ANOVA to analyse the main effects of N and P, plus their interaction (SAS, 1990).

Results and discussion

Data presented in Table 1 show that the number of tillers and plant height were significantly affected by N and P levels. Both characteristics were significantly increased with increasing N and P levels, but there were no differences between N150 or N200 under P100 level. The tallest plants (195 cm) were obtained by applying N150 or N200 and P100 compared to the control treatment height of 149 cm.

N and P fertilization significantly affected the herbage CP content. CP progressively increased with the increasing levels of applied N and P up to 200 kg N and 100 kg P ha^{-1}. The highest herbage CP content was obtained in the N200 and P100 treatment (8.5%), which was higher than the control treatment (6.6%), but statistically similar to N150 and P100 level (8.4%). The higher CP content at higher N and P levels was mainly due to the structural role of N in building up amino acids (Tegami Neto and Mello, 2007). Progressive increases in the CP contents of GKG swards with the increasing N and P rates were also reported by Hasyim *et al.* (2007) and Ruviaro *et al.* (2008). The NxP interaction was highly significant for the fresh herbage yield per plant but not for DM yield (Table 1). The highest fresh weight yield (1,719 g

Table 1. Effect of different N and P levels on the yield and some yield components of giant king grass.

	P0	P50	P100	Mean	P0	P50	P100	Mean	P0	P50	P100	Mean
	Tiller number ($plant^{-1}$)				Plant height (cm)				Crude protein content (%)			
N0	7.0	7.5	9.3	7.9	149	154	158	153	6.6	6.7	6.8	6.7
N50	8.3	8.8	9.8	8.9	157	164	175	165	7.0	7.5	7.4	7.3
N100	8.8	9.0	10.3	9.3	163	174	183	173	7.4	7.6	8.1	7.7
N150	9.3	9.3	11.3	9.9	175	180	195	183	7.5	7.8	8.4	7.9
N200	9.8	10.0	11.3	10.3	179	182	195	185	7.8	7.9	8.5	8.0
Mean	8.6	8.9	10.4	9.3	164	171	181	172	7.2	7.5	7.8	7.5
LSD[1] (0.01)	P:0.7 N:0.9 PxN:ns				P:3.4 N:4.3 PxN:7.5				P:0.1 N:0.2 PxN:0.3			
	Fresh herbage yield (g $plant^{-1}$)				DM yield (g $plant^{-1}$)				Dry root weight (g $plant^{-1}$)			
N0	765	987	1318	1023	155	225	320	234	120	130	135	128
N50	1061	1230	1480	1257	214	270	354	280	135	160	215	170
N100	1108	1303	1555	1322	222	287	356	288	215	220	315	250
N150	1139	1330	1719	1396	226	283	384	298	225	281	350	285
N200	1180	1353	1700	1411	230	279	364	291	270	300	343	305
Mean	1051	1241	1554	1282	210	270	357	279	193	218	272	228
LSD (0.01)	P:43 N:57 P×N:98				P:14 N:18 P×N:ns				P:4.9 N:6.4 P×N:11.1			

[1] LSD = least significant difference; ns: not significant.

plant^{-1}) was obtained using N150 and P100, whereas the lowest yield (765 g plant^{-1}) was recorded in the controls. N and P fertilization regimes also significantly (P<0.01) influenced grass DM yield.

The highest DM yield (384 g plant^{-1}) was expressed under the N150-P100 combination, consistent with established findings (Hasyim *et al.*, 2007; Ruviaro *et al.*, 2008). Analysis of herbage digestibility (not presented) demonstrated significant quality improvements at higher N and P levels. This is potentially attributable to physiological changes within the plants at differing fertilizer levels. Due to the number of tillers per plant increasing at higher applications of N and P, decreased stem production resulted in decreased herbage contents of neutral detergent fibre (NDF) and acid detergent lignin (ADL), giving rise to improvements in the digestibility value (Snijders *et al.*, 2011). Our findings are consistent with results reported by the Tessema and Baars (2006) and Snijders *et al.* (2011), whereby phenotype, N regime, management and cultivar all influenced above-ground yield and composition. The N×P interaction also significantly influenced root dry weight (Table 1). The highest root weight (350 g plant^{-1}) was obtained from N150 and P100 levels, whereas the lowest root weight (120 g plant^{-1}) was recorded in control pot. In our study, root growth was improved when N and P were applied. These data verified earlier results of Wadi *et al.* (2003), who reported that P applications increased root growth in *Pennisetum*.

Conclusions

Applications of N and P have a positive effect on herbage yield, quality and root growth of giant king grass if other growing conditions allow. The combination of 150 kg N and 100 kg P ha^{-1} had the highest forage yield and forage quality for GKG under controlled conditions. However, in spite of these results, further research is required to ascertain the optimal level of fertilizer in giant king grass cultivation under field conditions.

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Yield and silage characteristics of *Pennisetum hybridum* as affected by plant densities

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Abstract

The aims of this study were to determine the growth potential of Giant king grass (GKG) under Mediterranean climate conditions, including the effect of plant density on growth and yield. The experimental field was located at Izmir, Turkey. Treatments consisted of four densities of GKG (D1: 57,143; D2: 28,571; D3: 19,048 and D4: 14,286 plants ha^{-1}) with three replicates per treatment in a randomized block design. Sets of GKG were planted in mid-June of 2010 and allowed to grow for 5 full growing seasons. The mean result of 5 years indicated that there were significant effects of plant density on the dry matter yield and other yield characteristics (number of tillers, plant height, crude protein content) of GKG but not on silage pH. Density D2 was the most successful planting density of GKG regarding dry matter (32.6 t ha^{-1}) and crude protein yield. It is therefore the recommended sowing density of GKG within regions with Mediterranean-type climates.

Keywords: giant king grass, plant densities, yield

Introduction

Giant king grass (*Pennisetum hybridum*) as a field crop is a perennial grass and native to Africa (Hanna *et al.,* 1984), and has been the most promising and high yielding fodder grass, surpassing most other tropical grass. GKG has the potential to grow with low input, including low levels of irrigation and to tolerate a variety of biotic and abiotic stresses as compared to maize. Because it is perennial replanting every year is not required ('t Mannetje, 1992). Numerous field experiments carried out on *Pennisetum* genus indicated that yield and forage quality of the crop depends on cultivar, plant density, cutting interval and harvesting purpose. Optimum plant density is an important factor in maximizing yields of tall grasses such as *Pennisetum* or *Miscanthus* (Tegami and Mello, 2007; Zewdu, 2008). GKG is a recently introduced plant in Turkey. Therefore, knowledge of the production of GKG as crop species is important not only in Turkey, but also in southern part of Greece, Italy and Spain. Nevertheless, information on the productivity and quality of GKG under intensive farming management in Mediterranean environment is not well documented (Geren, 2014). The objective of this research was to evaluate the influence of different planting densities on yield and silage quality parameters of GKG under irrigated conditions of Mediterranean climate.

Materials and methods

The experiment was carried out during 5 growing seasons (2010-2014) at Bornova experimental fields of Agricultural Faculty of Ege University, Izmir, Turkey, at about 20 m above sea level under a typical Mediterranean climate. The cultivar of GKG used was 'Paraíso'. The experiment was carried out with a randomized complete block design with three replications; four plant spacing 70 cm among the rows and 25, 50, 75 and 100 cm within the rows (D_1: 57,143, D_2: 28,571, D_3: 19,048 and D_4: 14,286 plants ha^{-1}, respectively) were tested. Each plot was consisted of 4 rows with 5 m length (14 m^2). The grasses were grown from stem cuttings (sets) having 4 nodes and 40-60 cm length taken from a basal part of *P. hybridum* rootstock on 15 June, 2010. Before planting, the additional leaves of the setts were trimmed and dipped at 4,000 mg kg^{-1} indole-3-butyric acid (IBA) concentrations of hormone for 3-4 seconds up to 10-15 cm height. The recommended rate of 90 kg N ha^{-1} was applied to all plots in two equal rates during the establishment year (Ruviaro *et al.*, 2008). Half a dose of nitrogen (urea) fertiliser was applied before planting and the rest of nitrogen (ammonium sulphate) was applied when the plants were 50-60 cm high. All plots were fertilised using 100 kg ha^{-1} P_2O_5 before planting. In the following years, the same amount of N fertilization was applied in mid-April (in addition to phosphorus), as ammonium sulphate, and the other half at the beginning of stem elongation, as ammonium nitrate. Drip irrigation system was installed on the field during the establishment and growing seasons. Plots were harvested once a year at the beginning of November. Tiller density was also determined each year by counting the plants in the mid two rows in each plot at the same day of cutting. Forage from 3 m^2 in each plot was cut at 15-20 cm above ground level and fresh weight was recorded. Samples of 250 g were chopped and wilted and vacuum-packed in polythene bags with addition of 0.5% salt. The vacuum bag silos were kept in storage without light for 60 days for anaerobic fermentation. pH value of matured silage samples was also determined. Crude protein (CP) was calculated by multiplying the Kjeldahl N concentration by 6.25. Neutral detergent fibre (NDF) and acid detergent fibre (ADF) concentrations of silage were measured by Ankom Technology (Macedon, NY, USA) to determine the relative feed value (RFV) (Trotter and Johnson, 1992). All data were statistically analysed by analysis of variance using SAS (1998).

Results and discussion

Field studies were started in mid-June with high air temperature and satisfactory moisture levels supported by drip irrigation and owing to IBA, therefore, stands were excellent. No winter injury on the crops was detected during the experimental years. The results are summarized in Table 1.

Table 1. Effect of different plant densities on the yield and some yield components of giant king grass.

Density[1]	2010	2011	2012	2013	2014	Mean	2010	2011	2012	2013	2014	Mean
	Tiller number (m^{-2})						Plant height (cm)					
D_1	35	232	259	270	294	218	212	327	409	388	395	346
D_2	40	259	292	294	304	238	229	352	410	394	406	358
D_3	45	271	295	296	301	242	257	374	420	403	404	372
D_4	54	287	302	303	302	250	278	382	426	404	401	378
Mean	44	262	287	291	301	237	244	359	416	397	401	364
LSD[2] (0.05)	Y:10 D:12 Y×D:ns						Y:11 D:14 Y×D:17					
	Dry matter yield (t ha^{-1})						Silage pH					
D_1	15.6	32.2	35.5	36.9	38.0	31.6	3.86	3.77	3.65	3.65	3.63	3.71
D_2	18.4	35.1	37.5	39.4	39.8	34.0	3.80	3.59	3.63	3.64	3.62	3.66
D_3	17.7	33.1	35.5	37.8	38.6	32.5	3.78	3.57	3.61	3.62	3.65	3.65
D_4	17.7	31.6	33.4	35.2	37.3	31.0	3.60	3.54	3.60	3.61	3.63	3.60
Mean	17.3	33.0	35.5	37.3	38.4	32.3	3.76	3.62	3.62	3.63	3.63	3.65
LSD (0.05)	Y:1.2 D:1.4 Y×D:ns						Y:ns D:ns Y×D:ns					
	Crude protein content (%)						Relative forage value					
D_1	7.7	7.0	6.5	6.3	6.4	6.8	125	96	91	89	87	97
D_2	7.2	6.4	6.3	6.2	6.3	6.5	118	94	89	88	87	95
D_3	6.5	5.7	5.8	6.1	6.2	6.0	111	89	87	86	85	92
D_4	6.4	5.6	5.7	6.0	6.0	5.9	105	86	85	85	84	89
Mean	6.9	6.2	6.1	6.2	6.2	6.3	115	91	88	87	86	93
LSD (0.05)	Y:0.3 D:0.3 Y×D:ns						Y:3 D:4 Y×D:ns					

[1] D_1: 57,143, D_2: 28,571, D_3: 19,048 and D_4: 14,286 plants ha^{-1}.

[2] LSD = least significant difference; Y = year, D = density, Y×D = interaction, ns = not significant.

There were statistically significant differences among GKG plant densities regarding average number of tillers per square meter. Number of tillers (TN) in the last 4 years was approximately 6-7 times higher than the TN in the first production year. The average of 5 year results indicated the decreasing plant density from D_1 to D_4 increased the number of tillers per square meter, but there was no significant difference between D_2 and D_3. The plant height of GKG was affected by Y×D interaction. The five year average showed that the plant height of GKG increased noticeably by decreasing plant density most probably due to thicker stem plants developed at wider rows with less competition compared to narrow spacing. Zewdu (2008) informed that there was no significant effect on plant height of *P. purpureum* due to the plant density during the initial year however, plant height was significantly affected by plant density in the following year, and plants at lower plant densities were taller than at the higher densities.

In our study, there were statistically significant differences among plant densities regarding dry matter (DM) yield per hectare. According to the five year results, the maximum DM yields occurred at D_2. This result suggests that the number of tillers per area, plant height and stem thickness can cause a strong competition among the crops and depress the yield components of individual plants in lower or wider densities. Nevertheless, since the numbers of tillers per area in sparsely populated stands (D_3-D_4) were quite higher than the other densities which were densely populated stands (D_1-D_2), it was also suggested that DM yields were the highest at D_2. Even thought, number of tillers per area and plant height increased with decreasing rate of plant density. Plant density did not show any significant effect on the silage pH of GKG silage in the study. The average indicated that plant density affected CP content of GKG significantly, and densely populated stands gave higher CP content compared to sparsely populated stands. Higher plant densities (D_1 and D_2) had higher RFVs on average, whereas RFV at lower plant densities (D_4 and D_3) were lower, however the differences between them were not significant.

Conclusions

Within the Mediterranean coastal part of Turkey, giant king grass is a promising perennial forage crop with a high level of adaptability, forage yield and quality pecularities. The results of our multi-year study with four different plant densities showed that based on DM and CP yields, the planting of GKG using 28,571 (70×50 cm) plants ha^{-1} should be recommended in the regions with Mediterranean-type climates and in similar agro-ecological conditions.

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Forage yield and nutritional values of *Pennisetum purpureum* as affected by cutting height

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Abstract

A pot-experiment was conducted to investigate the effect of 4 different cutting heights (5, 10, 15 and 20 cm) on forage yield, and nutritional values of Napier grass (*Pennisetum purpureum*). Characteristics such as plant height, number of tillers, dry matter content and yield, content of crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF) were measured in the study. There were 4 replicate pots for each treatment. Results indicated that cutting height affected the fresh and dry matter yields and nutritive values of Napier grass. Total dry matter yield was significantly higher at a 15 cm cutting height than at 5 or 20 cm cutting height. There were significant differences in CP, NDF and ADF content among the treatments, CP ranging from 11.6 to 12.9%. Cutting height of 15 cm above ground level could be the optimal level for harvesting Napier grass in a coastal part of the Aegean region under Mediterranean climate.

Keywords: napier grass, cutting height, forage yield, nutritive composition.

Introduction

Napier grass (*Pennisetum purpureum*) also known as 'Elephant grass' is a perennial forage grass with a high growth rate, high productivity and good nutritive value and it is mostly used for 'cut and carry' system in the tropical and sub-tropical areas of the world (Wadi *et al.*, 2004). It can withstand repeated cutting and regrows rapidly, producing a high biomass that is palatable in the leafy stage (Van de Wouw *et al.*, 1999). The various studies on cutting Napier grass reveal that both the choice of a cutting interval and height of cutting are crucial to their performance and it has been found that the main factor affecting growth, yield and persistence of swards is the defoliation intensity. The harvesting intensity affects the regrowth capacity and determines the herbage quality (Singh *et al.*, 2013). However, herbage yield is affected both by plant species, cultivar and the environmental conditions. Cutting near the ground level increases total yield over a short period compared to more elevated cutting heights. Plants are adversely affected by frequent cutting because they lose the growing point and reduce carbohydrate storage in the subterranean stem of stubble tillers. Wadi *et al.* (2004) found that Napier grass cut at a 30 cm height was superior to the grass cut at a 0-cm height. Lounglawan *et al.* (2014) reported that cutting height at 5, 10 and 15 cm had no significant effect on dry matter (DM) and nutrient yields of king Napier grass. Tekletsadik *et al.* (2004) recommended a 20 cm cutting height as the optimum cutting height for dwarf Napier grass. Aganga *et al.* (2005) found that young and immature Napier grass cut at 50 cm height was highly digestible but as maturity increased, yield also increased, but quality decreased. This study was designed to determine the effect of different cutting heights on the forage yield and nutritional value of Napier grass under outdoor conditions.

Materials and methods

Dwarf variety 'Mott' of Napier grass (*P. purpureum*) was grown outdoors in a pot experiment from May to November in 2015 in an experimental area of Field Crops Department, the Faculty of Agriculture, University of Ege, Izmir/Turkey. Pots (24 cm diameter, 40 cm depth) were filled with commercial field soil (16 kg). The physical properties of soil used were: 80.2% sand, 18.0% silt and 1.8% clay; and chemical properties were: pH 5.83, salt 0.03%, organic matter 2.27%, total N 0.092%, available $CaCO_3$, P, K were 1,300 μg kg^{-1}, 2.54 μg kg^{-1} and 40 μg kg^{-1}, respectively. Four different cutting heights (5, 10, 15 and 20 cm) were tested. The experiment was arranged by a completely randomized block design with 4 replications. The seeds were sown in a plastic pot tray filled with a mixture of sand, manure and mulch (1:1:1) at the beginning of March 2015 under greenhouse conditions, at 25 ±1 °C. When the seedlings of Napier grass reached 10-15 cm in plant height, they were transferred to the experimental pots (23 May 2015). For all pots, two equal rates of 200 kg N ha^{-1} were applied. The first rate was added before planting to the pots (plus 100 kg ha^{-1} P_2O_5 and 100 kg ha^{-1} K_2O) and the other N was added after the first cutting practices (Singh *et al.*, 2013). Moisture was maintained by watering plants every other day with 250 or 500 ml of tap water according to moisture content of the soil. The crops were harvested at the beginning of the blooming stage (9 July) under different stubble heights above mentioned. After each cut, plant height was measured, number of tillers per plant was determined by counting the plants in the pot, fresh forage was weighed and subsamples were taken for determination of DM (in an oven dried at 65 °C for 3 days). Crude protein (CP) (Kjeldahl $N_{\%} \times 6.25$) was calculated, while acid detergent fibre (ADF) and neutral detergent fibre (NDF) were analysed by the method of detergent analysis (Goering and Van Soest, 1970). At the end of the experiment, the soil was gently washed from the roots, and the roots were oven dried at 65 °C for 3 days. All data were statistically analysed using analysis of variance (ANOVA) with the Statistical Analysis System (SAS, 1990). If ANOVA indicated differences between treatment means, a least significant difference test (0.01) was performed to separate them.

Results and discussion

Data presented in Table 1 show that with increasing cutting height up to 15 cm, plant height (PH), leaf rate (LR), number of tillers (NOT), dry matter yield (DMY) and dry root weight (DRW) increased, while NDF and ADF contents decreased.

Protein content increased with higher cutting heights and the highest protein content was obtained at a cutting height of 20 cm, which is in contrast to the results obtained by Wijitphan *et al.* (2009). They found no significant differences among CP contents when applying different cutting heights. In our research, the lowest DMY, NOT, LR, CP, ADF, NDF and DRW were obtained by a cutting height of 5 cm. Harvest height had a significant (P<0.01) impact on measured parameters, with increase in main parameters when cutting was up at a height of 15 cm. This study verified earlier results that Napier grass can be superior with higher cutting height (Wadi *et al.*, 2004, Wijitphan *et al.*, 2009), but it differs from the results of Lounglawan *et al.* (2014) who showed that cuttings at 5, 10 and 15 cm had no significant effect on dry matter and nutrient yield. Based on this research, NDF and ADF concentrations significantly decreased with the advance of the cutting height. Similar results of ADF and NDF were obtained by Kozloski *et al.* (2003) at different cutting ages of Napier grass (ADF 330-353 g kg^{-1}, NDF

Table 1. Effect of different cutting heights on the yield and nutritional values of *Pennisetum purpureum*.[1,2]

Cutting height	TNC	Plant height (cm)	Leaf rate (%)	Number of tillers ($plant^{-1}$)	DM yield (g $plant^{-1}$)	CP (g kg^{-1})	NDF (g kg^{-1})	ADF (g kg^{-1})	Dry root weight (g $plant^{-1}$)
5 cm	3	82.1 c	48.1 c	20.9 d	373 c	116 c	611 a	411 a	47 c
10 cm	4	86.8 b	58.4 a	42.0 b	627 a	123 b	598 b	392 b	155 a
15 cm	4	92.1 a	59.7 a	46.1 a	654 a	124 b	554 c	376 c	160 a
20 cm	4	76.9 d	51.8 b	30.1 c	421 b	129 a	549 c	371 c	118 b
Mean		84.5	54.5	34.8	519	123	578	387	120

[1] TNC = total number of cuttings during the growing season; DM = dry matter; CP = crude protein; NDF = neutral detergent fibre; ADF = acid detergent fibre.

[2] Means followed by different letters are significantly different at the 0.01 level of probability.

608-648 g kg^{-1}). The findings from this study suggested a strong correlation between dry root weight and dry matter yield.

Conclusions

Based on this research, it can be concluded that the cutting height may affect the forage dry matter yield and nutritive value of Napier grass. Results suggested that the high level of cutting can improve not only dry matter yield, but also forage quality of Napier grass. The effect of greater cutting heights tended to reduce ADF and NDF. These findings have important implications for the use of Napier grass as a livestock feed. A cutting height of 15 cm above ground level could be the optimal level for harvesting Napier grass. However, field experiments in the coastal part of the Aegean region under Mediterranean climate still need to confirm practical recommendations to farmers.

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Effect of cutting intervals on the forage yield and some yield characteristics of Napier grass

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Abstract

This study was conducted to investigate the effect of 9 different cutting intervals (30-45-60-75-90-105-120-135-150 days) on the fresh forage yield and chemical composition of Napier grass (*Pennisetum purpureum*). Some characteristics such as plant height, number of tillers, dry matter (DM) content and yield, content of crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF) were tested in the study. There were 4 replicate pots for each treatment. Results indicated that cutting intervals affected DM yields and nutritive values of Napier grass. As the inter-cutting interval increases the percentages of leaf in the harvested material declined and stem percentage increased, with a concomitant decrease in CP concentration and increases in cell wall contents. Plant DM yield significantly increased with plant age at cutting, with age at cutting of 60 days yielding highest, followed by 75 days in a coastal part of the Aegean region under Mediterranean climate.

Keywords: Napier grass, cutting interval, forage yield, nutritive composition

Introduction

Napier grass (*Pennisetum purpureum*) also known as 'Elephant grass' is a perennial forage grass with a high growth rate, high productivity and good nutritive value and it is mostly used for 'cut and carry' system over the tropical and sub-tropical areas of the world (Wadi *et al.*, 2004). Napier grass is very palatable to animals at the early leafy stage, but not much liked after stem development. Frequency of defoliation after crop establishment is the common agronomic factors, which affect growth characteristics, dry matter (DM) yield and nutritional quality of Napier grass (Lounglawan *et al.*, 2014). The harvesting frequency or interval differs by climatic conditions but should coincide with the timing of high forage quality and the duration between two cuttings depending on the time needed by the plant to regrow (Wadi *et al.*, 2004). The DM yield is higher if the plant is permitted to develop stems, but it decreases leaf blade to stem ratio, reducing the forage quality as a result of lowered protein content and digestibility. The regrowth after cutting practices is one of the most important physiological processes and it determines the herbage quality. Wijitphan *et al.* (2009) reported that DM yield of Napier grass was increased by extending the intervals of cutting from 4 to 7 weeks. In another study, Mohammad *et al.* (1988) obtained highest DM yield on 3.3 t ha^{-1} from Napier grass clipped at 8 week interval. However, before recommending Napier grass as a forage crop to farmers, it is essential to understand and determine the relationships between growth characteristics, DM yield, nutritional quality and management practices such as frequency of defoliation to maintain growth of Napier grass. This study was designed to determine the effect of different cutting intervals on the forage yield, yield components and nutritional values of Napier grass under controlled conditions.

Materials and methods

The dwarf variety 'Mott' of Napier grass (*P. purpureum*) was grown outdoors, in a pot of 24 cm diameter and 40 cm depth filled with commercial field soil (16 kg), in an experimental area of Field Crops Department, the Faculty of Agriculture, University of Ege, Izmir/Turkey for 8 months in 2015. The physical properties of soil used were: 80.2% sand,18.0% silt, 1.8% clay; and chemical properties were: pH

5.83, salt 0.03%, organic matter 2.27%, total N 0.092%, available $CaCO_3$, P, K were, 1,300 μg kg^{-1}, 2.54 μg $kg^{-1}$and 40 μg kg^{-1}, respectively. Nine different cutting intervals (30, 45, 60, 75, 90, 105, 120, 135 and 150 days) were tested on Napier grass. The experiment was arranged by a completely randomized block design with 4 replications. The seeds were sown in a plastic pot tray filled with a mixture of sand, manure and mulch (1:1:1) at the beginning of March 2015 under greenhouse conditions, at 25±1 °C. When the seedling of Napier grass reached 10-15 cm in plant height, they were transferred to the experimental pots on 23 May, 2015. For all pots, in three equal rates of 150 kg N ha^{-1} were applied. The first 1/3 N (urea) rate was added before planting to the pots (plus 100 kg ha^{-1} P_2O_5 and 100 kg ha^{-1} K_2O) and the others N [$(NH_4)_2SO_4$] were added 60 and 90 days after transferring of the crops (Wadi *et al.*, 2004). Moisture was maintained by watering plants every other day with 250 or 500 ml of tap water according to moisture content of the soil. The grass was clipped manually using a hand tool-sickle with 10 cm stubble height at the 9 cutting frequencies 30 to 150 days after the crop establishment. After each cut, plant height was measured; number of tillers per plant was also determined by counting the plants in the pot and the fresh forage was weighed and subsamples were taken for determination of DM content (in an oven dried at 65 °C for 3 days), crude protein (CP) (Kjeldahl $N_{\%}$ ×6.25) was calculated, while acid detergent fibre (ADF) and neutral detergent fibre (NDF) were analysed by the method of detergent analysis (Goering and Van Soest, 1970). At the end of the experiment, the soil was gently washed from the roots, and the roots were oven dried at 65 °C for 3 days. All data were statistically analysed using analysis of variance (ANOVA) with the Statistical Analysis System (SAS, 1990). If ANOVA indicated differences between treatment means, a least significant difference value (0.01) is given.

Results and discussion

We obtained '5, 3+, 2+, 2, 1+, 1+, 1+, 1+, 1' number of cuts for intervals 30, 45, 60, 75, 90, 105, 120, 135 and 150 d, respectively, in our study. Data is presented in Table 1 as total (yield) or average values of cuttings. There was a significant (P<0.01) effect of cutting interval (frequency) on all growth, yield and chemical characteristics of Napier grass (Table 1).

Plant height increased as the cutting frequency decreased (30 d [5 times] →150 d [once]), whereas leaf ratio and number of tillers per plant decreased. The 30 d interval had lowest plant height (52.6 cm) while the 135 d gave the highest plants (116.5 cm). The number of tillers and leaf ratio per plant ranged from 52.7 to 21.6 $plant^{-1}$ and 66.1 to 52.7%, respectively. Clavero (1997) reported that clipping after 42 or 56

Table 1. Effect of different cutting intervals on the yield and nutritional values of *Pennisetum purpureum.*[1]

Cutting interval (days)	Plant height (cm)	Leaf rate (%)	Number of tillers (pot^{-1})	DM yield (g pot^{-1})	Dry root weight (g pot^{-1})	CP (g kg^{-1})	NDF (g kg^{-1})	ADF (g kg^{-1})
30	52.6	66.1	52.7	344	128.3	131	494	337
45	76.5	64.3	45.4	473	130.0	129	511	341
60	77.8	63.5	39.4	694	181.7	124	517	352
75	78.4	62.9	38.7	650	220.0	121	524	368
90	82.7	59.7	33.1	586	161.7	120	547	379
105	100.4	58.4	32.5	554	151.7	118	568	385
120	102.7	55.9	30.5	509	148.3	110	581	392
135	116.5	55.4	29.6	509	135.0	104	593	402
150	114.6	52.7	21.6	483	145.0	103	608	408
Mean	89.1	59.9	35.9	534	155.7	118	549	374
LSD	7.5	1.7	4.2	70	12.5	4	6	5

[1] DM = dry matter; CP = crude protein; NDF = neutral detergent fibre; ADF = acid detergent fibre; LSD = least significant difference.

days gave maximum tillers (45 and 34 five plant^{-1}) with 20 cm stubble, which contributed to regrowth and stand density. The leaf ratio per plant decreased with increasing days between cuttings. Interval of 150 d was the inadvisable in terms of number of tillers and leaf ratio per plant. The decreasing trend, observed in leaf-to-stem ratio as the growth period increased, was in agreement with the report of Butt *et al.* (1993), who indicated that the reduction in leaf proportion and an increase in stem fraction of Napier grass following delayed harvest occurred because of maturity.

Cutting interval had a significant (P<0.01) effect on DM yield and dry root weight of Napier grass. The DM yield increased as the frequency of cutting decreased from 30 to 60 d (344 g and 694 g pot^{-1}, respectively), but no significant differences between 60 d and 75 d (650 g pot^{-1}), however after 90 day, DM yield decreased most probably due to the regrowth period for additional cutting in our study. Dry root weight increased from 30 to 75 d, whereupon it decreased. All chemical components such as CP, NDF and ADF were significantly (P<0.01) affected by frequency of defoliations. CP contents decreased with decreasing frequency of cutting, whereas NDF and ADF contents showed an increasing trend with a decrease in defoliation frequency (Table 1). CP content declined from 131 to 103 g kg^{-1}; however NDF and ADF contents raised from 494 to 608 g kg^{-1} and 337 to 408 g kg^{-1}, respectively, with decreasing frequency of defoliation from 30 to 150 d after Napier grass establishment. It is widely reported that CP content decreases as cutting interval increases (Tessema *et al.*, 2010). In contrast, NDF and ADF increase with increasing days of defoliation (Wangchuk *et al.*, 2015).

Conclusions

Considering overall results of our pot experiment, it could be concluded that cutting interval significantly affected the plant growth, forage yield and quality. Cutting at 60 or 75 daily intervals seems to provide maximum DM yield with acceptable leaf ratio and nutritional quality. However, field experiments with feeding and digestion studies are needed to confirm the results.

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How much potassium for silage maize on light sandy soils?

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Abstract

Improving the eco-efficiency of silage maize production requires a nutrient supply fine-tuned to the crop demand. The impact of potassium supply on silage maize dry matter yield was studied in a multi-year field trial on a light sandy soil in northern Germany. A potassium supply of 83 kg K ha^{-1} was sufficient for maximising dry matter yield, while K demand was higher for maximum K yield and a zero K balance. An equation was derived to quantify the critical K content as a function of biomass, which may provide a tool for evaluating the K fertiliser management after validation.

Keywords: critical K concentration, fertilisation, soil K content, yield, *Zea mays* L.

Introduction

Potassium is involved in many physiological processes and has direct consequences on crop yield, e.g. via its impact on crop water relations and photosynthesis. In Schleswig-Holstein, northern Germany, a region with high-output silage maize production, an increase in the proportion of arable soils with low K status has been identified in monitoring by the Agricultural Chamber, indicating that the supply of basic nutrients obviously has decreased. This can be attributed to changes in fertiliser price and a decline in the K content of cattle slurry observed over recent years. The objectives of the current study therefore were (1) to analyse the silage maize yield response to K fertilisation on a typical light sandy soil, and (2) to derive a critical K content as an indicator of the maize K status.

Materials and methods

The study was based on a continuous maize experiment (9.4° E, 54.5° N) established as a randomised block design with 4 replicates in 2007 on a sandy soil (podzol, pH 4.8-5.3) in northern Germany. Data analysis included three years (2010-2012). Potassium supply to silage maize (*Zea mays* L cv. Salgado, early) was varied by four levels (0, 83, 166, and 249 kg K ha^{-1}), applied as potassium chloride before sowing at the beginning of April. All plots received a total N fertilisation of 140 kg N ha^{-1} (2010, 2012: Entec, 2011: urea), split into two dressings, i.e. a banded starter of 40 kg N ha^{-1} plus 100 kg N ha^{-1} one to two weeks after sowing. Similarly, phosphorus was given in two partial applications, i.e. 21.8 kg P ha^{-1} two to four weeks before sowing and 19.6 P ha^{-1} as banded starter. Further nutrients were applied according to good agricultural practice in order to avoid any nutrient deficiencies. The dynamics of biomass accumulation were recorded by five manual samplings (10 plants per plot) during the vegetation period and a final machine harvest at silage maturity. Sub-samples were dried at 58 °C and 105 °C. Potassium content was determined using a flame photometer (763 nm). Maize energy concentration was estimated by near-infrared spectrometry, based on GfE (2009) and Weißbach *et al.* (1996). An analysis of variance was conducted to determine the impact of K fertilisation and year on dry matter (DM) yield, K yield and K balance (K input minus K yield) using SAS 9.2 ProcMixed, where K fertilisation, year as well as their interaction were assumed fixed, and block as random. Multiple comparisons of means were performed by t-test and Bonferroni-Holm adjustment or Tukey-test. The critical K content was derived based on all sampling dates according to Herrmann and Taube (2004) using an exponential function for quantifying the relation between DM yield and critical K content. Changes in soil K content were also monitored by regular soil sampling.

Results and discussion

The results revealed a significant impact of K fertilisation and of year on silage maize DM yield. Compared to the control (0 kg K ha^{-1}), yield increased significantly by 5.0, 5.7 and 5.8 t DM ha^{-1} in the fertilised treatments (Figure 1A), but no significant differences were detected among them. Thus, maximum yield was already achieved with a K input of 83 kg K ha^{-1}. However, it cannot be concluded that 83 kg K ha^{-1} is sufficient for the long-term cultivation. Maize energy concentration (MJ NEL kg^{-1} DM) and energy yield (GJ NEL ha^{-1}) showed the same pattern as DM yield (data not presented). The K removed by harvest (K yield) ranged between 41 kg K ha^{-1} (control, 2012) and 184 kg K ha^{-1} (highest K level, 2011) and was affected by the interaction of K fertilisation and year (Figure 1B). In contrast to DM yield, a K input of 166 (2011, 2012) or 249 kg K ha^{-1} (2010) was required for maximum K yield, indicating luxury K consumption. Similar to K yield, the K balance was influenced by the interaction of K fertilisation and year (Fig. 1C). As expected, the K balance increased continuously with K supply. A K balance close to zero can be expected for a K fertiliser input between 83 and 166 kg K ha^{-1}.

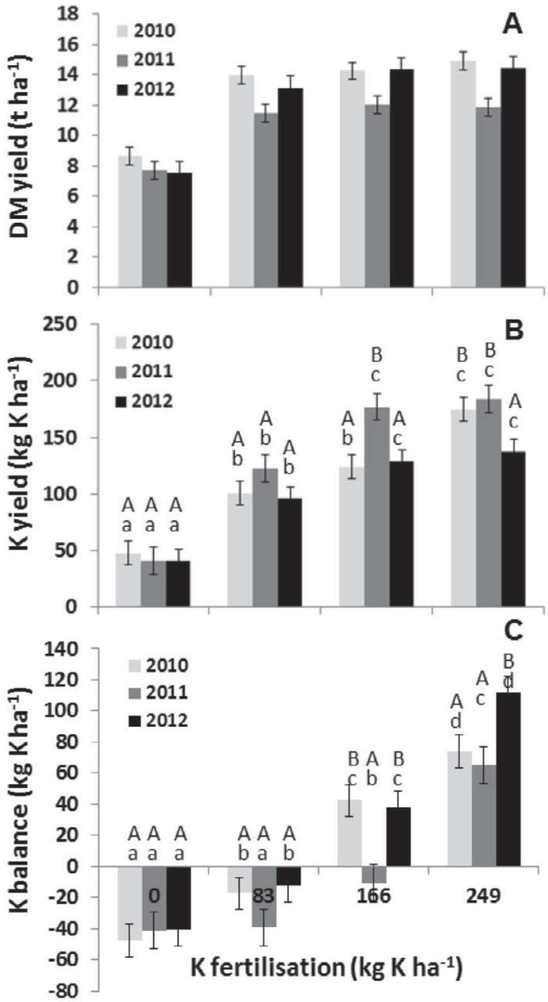

Figure 1. Dry matter yield, K yield and K balance of silage maize as affected by K fertilisation and year. Lower case letters indicate significant differences among K fertiliser levels within a given year, capital letters mark significant differences among years within a given K level.

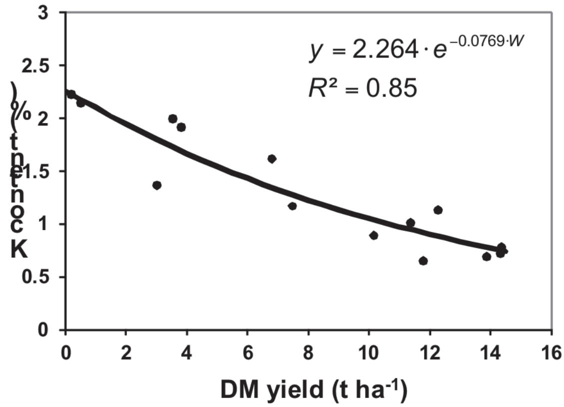

Figure 2. Relationship between maize critical K content (%) and dry matter yield (t dry matter (DM) ha^{-1}). W = dry matter yield in t DM ha^{-1}).

At the beginning of the field experiment (2007), soil K content was very low (2.5 mg K (100 g)$^{-1}$ soil, determined by the double-lactate method, corresponding to soil K class A). The control and 83 kg K treatments revealed marginal changes in K content and stayed in soil K class A over the whole experimental phase. The 166 and 249 kg K treatments resulted in increased soil K contents, achieving class C (9.1 mg K (100 g)$^{-1}$ soil; optimal K status) in 2012. To exploit the silage maize yield potential on light sandy soils, it is obviously not required to increase the soil K status to a level (class C), which is regarded optimal according to the official fertiliser recommendations. Instead, K supply should be adjusted to crop K removal, which would also reduce the risk of K leaching (Kayser *et al.*, 2012).

A function quantifying the critical K content of maize could be successfully derived (Fig. 2). For a silage maize yield of 13 t DM ha^{-1}, which is the average yield at farm level in the federal state of Schleswig-Holstein, a plant K content of 0.83% thus can be regarded as sufficient. Older studies often provide higher critical values. Leigh *et al.* (1984), for instance, reported the photosynthesis rate of maize leaves to be substantially reduced for K contents below 1.1 to 1.5%. For grain maize, the authors regarded a K content of 1.3% necessary for achieving 90% of the maximum yield. An impact of N supply or plant density on K content can be largely excluded, apart from severe K deficiency (Ciampitti *et al.*, 2013). Differences in K uptake efficiency among hybrids are mainly dependent on differences in root architecture (Feil *et al.*, 1992; Cao *et al.*, 2007). For north-western Europe, however, no comprehensive studies on the impact of maize hybrid on K use efficiency are available.

Conclusions

Silage maize has a high K-uptake potential. A high K supply, however, is not necessarily converted into biomass. For light soils, increasing the soil K status to a level regarded as optimal according to the official fertiliser recommendations, seems inappropriate. Instead, the fertiliser recommendation should be adjusted to the K removal by harvest. The critical K curve, quantifying the critical K content of maize as a function of shoot DM yield, may serve as a useful tool for evaluating the efficiency of K fertilisation. Before implementation in practice, however, the concept requires validation at other sites and with different maize hybrids.

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Yields and feed value of different fodder galega-grass mixtures

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Abstract

Fodder galega (*Galega orientalis* Lam.) is a forage legume that has been grown in Estonia for almost 44 years. Pure galega is known to be a persistent, high-yielding crop and rich in nutrients, in particular crude protein (CP). Galega is usually grown in mixtures with grass in order to optimize crop nutrient concentration, increase dry matter (DM) yield and improve fermentation properties. In the present study galega was grown in mixture either with reed canarygrass (*Phalaris arundinacea* L.) cv. Marathon, timothy (*Phleum pratense*) cv. Tika, red fescue (*Festuca rubra* L) cv. Kauni or festulolium (× *Festulolium*) cv. Hykor in three successive years (2013-2015). Three cuts were carried out during the growing season in all three years. The total dry matter yield varied from 7.6 to 13.7 t ha^{-1}. The CP concentration in the DM varied from 156 to 186 g kg^{-1}. Both DM-yield and CP concentration were dependent on year, mixture and fertilization. N fertilization compared to no fertilization favoured grass growth and reduced the role of galega in the sward.

Keywords: fodder galega, goat's rue, galega-grass mixtures, forage yield, fertilization

Introduction

Along with other legumes, fodder crops such as lucerne and clovers, goat's rue, i.e. fodder galega, has been grown in Estonia for ca. forty years. Galega is very persistent and has a high yield potential. Previous studies have revealed that the yields can possibly be 8.5 to 10.5 tons of dry matter and 1.7 to 1.8 tons of crude protein per hectare with CP concentration of 200-220 g kg^{-1} DM (Raig *et al.*, 2001). The nutritive value is highest when the 1st cut is taken at budding or at the beginning of flowering. In order to balance the need for nitrogen fertilizer with biological nitrogen fixation, it is favourable to grow galega in a mixture with grass. When choosing grasses for mixtures, the rate of phenological development of the species, its persistency and its nutritive value should be considered. Earlier results have shown that growing galega in mixtures with grasses improves the nutritive value and ensiling properties of the crop (Lättemäe *et al.,* 2005; Meripõld *et al.*, 2014).

Materials and methods

The experimental field was established in 2012 in Saku Estonia (local latitude N57° E25'). The study includes data from three years (2013-2015). The trial plots were established on a typical soddy-calcareous soil where the agrochemical indicators were as follows: pH_{KCl} 6.3 (ISO 10390); soil carbon content C_{org} 3.3% and concentration of lactate soluble P and K being 114 and 161 mg kg^{-1} respectively. Four galega-grass mixtures were tested. The galega cv. Gale (*Go*) was sown in binary mixtures with reed canarygrass cv. Marathon (*Pa*) (7 kg ha^{-1}), red fescue cv. Kauni *(Fr)* (10 kg seed ha^{-1}), timothy cv. Tika (*Pp*) (8 kg ha^{-1}) and festulolium cv. Hykor (*Fe*) (15 kg ha^{-1}) respectively. The sowing rate of Gale was 15 kg ha^{-1} in all mixtures. A pure fodder galega *(Go)* and festulolium (Fe) were included in the trial as controls. In order to increase competitiveness of grasses and the yield of the first cut, two N fertilization levels were applied: N0 and N 50 kg ha^{-1} (April or May). The crop was cut by a scythe, and then weighed and samples were taken for analyses. Prior to sampling the botanical composition of crop was determined. A three-cut system was used during harvest and three replicates of the plots of each treatment. The following data were collected in this experiment: dry matter yield (DM), crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF) and metabolisable energy (ME) contents. Effective temperatures over 5 °C for first cut in 2014 were 241 °C, 291 °C in 2013 and 178 °C in 2015. Effective temperatures of 2015

(April-May) were the lowest in the last ten years. The data were analysed by three-way ANOVA with mixture, N-fertilization and year as fixed factors. For the statistical analysis the Tukey-Kramer Honest Significant Difference (HSD) test was used via the software JMP 5.0.1.2 (SAS, 2002).

Results and discussion

The galega-grass mixtures returned high DM yields at both fertilization levels and in all experiment years. Yields varied from 7.6 to 13.7 t ha^{-1} (Table 1). The highest DM yield of the three experiment years was obtained from Gale-Hykor mixture supplied with N fertilizer. The yields were higher in 2015 and varied from 8.1 to 11.5 t ha^{-1}.

The botanical composition of the sward was influenced by the different climate conditions, the competitiveness of grasses and the application of N fertilizer. N fertilizer increased the proportion of grass in all mixtures (Figure 1). At the fertilization level N0 the red fescue cv. Kauni and festulolium cv. Hykor were less competitive (Figure 1). The highest proportion of grass appeared in mixtures with timothy cv.

Table 1. The dry matter yield (t ha^{-1}) of fodder galega-grass mixtures in 2013-2015.[1]

Species[2]	Mixture	2013		2014		2015	
		0 N	50 N	0 N	50 N	0 N	50 N
Go	Gale	10.5^{cdefg}	8.2^{ghi}	12.0^{abc}	10.7^{cdef}	9.9^{defgh}	9.6^{defgh}
Go/Pa	Gale/Marathon	7.6^{hij}	11.1^{bcde}	9.5^{defgh}	11.1^{bcde}	9.2^{efghi}	10.1^{defgh}
Go/Pp	Gale/Tika	8.7^{efghi}	10.3^{defgh}	9.4^{defgh}	13.2^{ab}	8.1^{ghi}	10.1^{defgh}
Go/Fr	Gale/Kauni	7.9^{hij}	10.3^{defgh}	11.4^{bcd}	11.4^{bcd}	9.6^{defgh}	10.0^{defgh}
Go/Fe	Gale/Hykor	8.2^{ghi}	12.1^{abc}	10.0^{defgh}	13.7^{a}	9.1^{efghi}	11.5^{bcd}
Fe	Hykor	5.5^{j}	10.2^{defgh}	6.2^{ij}	10.7^{cdef}	2.3^{k}	8.2^{ghi}
Level of significance for main effects and interactions							
Factor	Fertilization	Year	Mixture	Mixture × fertilization	Mixture × year	Mixture × fertilization × year	
P-value	<0.001	0.006	<0.001	<0.001	<0.001	<0.001	

[1] Different letters behind the mean values indicate significant differences ($P<0.05$).

[2] Go = *Galega orientalis*; Pa = *Phalaris arundinacea*; Pp = *Phleum pratense*; Fr = *festuca rubra*; Fe = *Festucolium*.

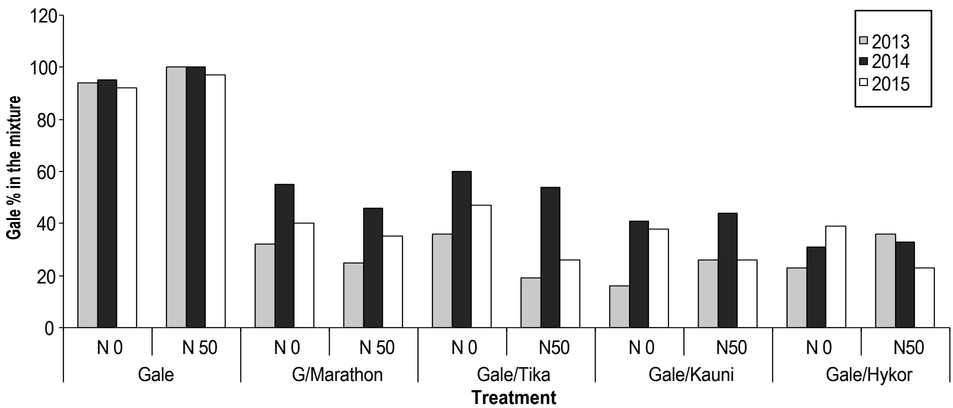

Figure 1. The botanical composition of galega-grass mixture of first cut in 2013-2015.

Table 2. The nutritive value of the fodder galega and grasses in mixtures and alone at first cut. Means for three years are given.[1]

Mixture	Fertilizer	CP (g kg^{-1})	NDF (g kg^{-1})	ADF (g kg^{-1})	ME (MJ kg^{-1})
Gale	0 N	238^{a}	461^{abc}	336^{a}	9.7^{a}
	50 N	222^{ab}	420^{bc}	320^{a}	9.9^{a}
Gale/Marathon	0 N	181^{abc}	475^{abc}	339^{a}	9.5^{a}
	50 N	184^{abc}	463^{abc}	325^{a}	9.8^{a}
Gale/Tika	0 N	171^{bc}	494^{a}	349^{a}	9.4^{a}
	50 N	180^{abc}	498^{a}	339^{a}	9.7^{a}
Gale/Kauni	0 N	175^{abc}	498^{a}	346^{a}	9.4^{a}
	50 N	181^{abc}	477^{abc}	321^{a}	9.9^{a}
Gale/Hykor	0 N	156^{c}	521^{a}	348^{a}	9.5^{a}
	50 N	186^{abc}	482^{ab}	331^{a}	9.9^{a}
Hykor	0 N	88^{d}	554^{a}	327^{a}	9.6^{a}
	50 N	143^{cd}	530^{a}	334^{a}	9.8^{a}
Level of significance for main effects and interactions (*P*-value)					
Fertilization		ns	ns	ns	ns
Mixture		0.002	0.001	ns	ns
Mixture × fertilization		0.004	0.038	ns	ns

[1] Within columns, means followed by different letters indicate significant differences ($P<0.05$). ns = not significant.

Tika and the reed canarygrass cv. Marathon at N50 fertilization level. When plots were fertilized, CP concentration and metabolisable energy (ME) increased but NDF and ADF decreased.

In general, the nutritive value of mixtures was mainly dependent upon fertilization level and 'Gale' proportion (Table 2). Lower CP in mixtures (156-181 g kg^{-1} DM) and metabolisable energy (ME) (9.4-9.5 MJ kg^{-1} DM) concentrations were found in treatments without N fertilizer. When plots were fertilized, CP (180-186 g kg^{-1} DM) concentration and ME (9.7-9.9 MJ kg^{-1} DM) increased. NDF and ADF concentrations in mixtures were lower in 2014 and 2015 than in 2013 as plants were less developed in 2015 due to fewer degree days over 5 °C.

Conclusions

The galega-grass mixtures maintained high yielding ability. The botanical composition of the sward was influenced by the different climate conditions, the competitiveness of grasses and the application of N fertilizer. High N fertilization rate favoured grass growth but reduced the role of galega in the sward. The higher yielding ability and similar higher metabolisable energy value were obtained in Gale-Hykor and Gale-Marathon mixtures. On the basis of these results, fertilization should be recommended in order to avoid grasses being lost from the pasture and to prevent N deficiency in the spring.

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Effects of inoculation with homolactic bacteria on losses and quality of wheat silage in different layers in bunker-silo

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Abstract

This aim of this study was to determine the effects of a homolactic inoculant containing *Lactobacillus plantarum* and *Pediococcus acidilactici* (1×10^5 cfu g^{-1} FM) on the fermentative osses and chemical composition of wheat silages across different layers of large-scale bunker silos. Whole crop wheat (40% of dry matter (DM)) was harvested at soft dough stage and packed into two bunker silos (30 t per silo). At ensiling, two treatments (control vs inoculant) and 3 silo layers (bottom= 0 to 40 cm vs middle= 40 to 80 cm vs top= 80 to 120 cm layer) were factorially arranged, with 12 replicates per layer. Chemical composition was determined by standard wet chemistry methods, whereas silage density was measured with a 5 cm diameter corer. Dry matter loss during storage was measured using tracer bags containing 1.5 kg of fresh chopped forage. As expected, silage located at the upper layer had lower DM density and higher DM loss. On the other hand, the homolactic inoculant decreased silage pH and DM loss in all silo layers. Inoculated silage had lower concentrations of ash, crude protein and ether extract compared with the control silage. Poorer preservation in the top layer reflected in lower *in vitro* DM digestibility. In conclusion, the homolactic inoculant reduced the DM loss during anaerobic storage of wheat silage.

Keywords: bacterial inoculant, *Lactobacillus plantarum,* wheat silage

Introduction

Most studies on silage inoculants evaluate these products in laboratory silos. However, it is important to validate those results in large-scale silos, focusing on quality differences that may occur in different silo layers. Additionally, nutrient losses depending on silage preservation directly affects dry matter (DM) recovery, quality and costs. Thus, the aim of this study was to determine the effects of a homolactic inoculant containing *Lactobacillus plantarum* and *Pediococcus acidilactici* on the fermentative losses and chemical composition of wheat silages in different layers of large-scale bunker silos.

Materials and methods

The experiment was carried at the State University of Middle-West and State University of Maringá, Paraná, Brazil. Wheat (cvBRS Umbu) was sown in 3 hectares, using a row spacing of 0.17 m and a stand density of 220 seeds m^{-2}. Whole crop wheat was directly harvested at soft dough stage (40% DM) with a pull-type forage harvester and stored into two bunker silos (30 t per silo, 3.5 m width × 11 m length × 1.2 m height). At ensiling, two treatments (control vs inoculant) and 3 silo layers (bottom = 0 to 40 cm vs middle = 40 to 80 cm vs top = 80 to 120 cm layer) were factorially arranged, with 12 replicates per layer (one silo per silage type). The inoculant contained *Lactobacillus plantarum* and *Pediococcus acidilactici* and was applied at a theoretical rate of 1×10^5cfu g^{-1} of forage (diluted in water, 4 l t^{-1}). During ensiling, twelve tracer bags (nylon, 12×50 cm, 85 μm pore size) containing 1.5 kg of treated forage (with or without inoculant) were allocated throughout each silo layer. Silos were filled simultaneously (every second load for each silo) in one working day, packed with a tractor and sealed with polyethylene film (black-on-white, 200 μm). The tracer bags were recovered during silage unloading (~14 cm d^{-1}) and used to determine DM loss. Chemical composition was determined by standard wet chemistry methods, whereas silage density was measured with a 5 cm diameter corer. Organic acids and ethanol were analysed by gas chromatography, whereas ammonia nitrogen ($N\text{-}NH_3$ in total N) was determined according to

Chaney and Marbach (1962). The *in vitro* DM digestibility (IVDMD) was measured using the DAISY^{II} method (Holden, 1999). Data collected for each variable were subjected to variance analysis (GLM procedure, SAS Institute Inc., Cary, NC), and differences between means were analysed by Tukey test at 5%.

Results and discussion

Silage density is an important physical parameter that determines silage porosity and its potential deterioration during both fermentation and feedout phases. In this study, losses in large-scale silos were related to silo layer and, in turn, silage density (Table 1). Therefore, silo layer affected DM content and IVDMD. The bottom layer had lower DM content (P<0.05), which shows moisture accumulation in this part of the silo. Wheat silage without inoculant showed higher ash content in middle and bottom layers, whereas the inoculated silage showed lower ash concentration in the middle layer. Weinberg *et al.* (2010) also observed variations in ash levels, which were attributed to the occurrence of fermentative losses in wheat silages. In addition, Kung Jr. (2013) observed that there is greater microbial activity in surface layers, which increases nutrient losses. In the present study, the changes in pH and fermentation profile (Table 2) indicate that homofermentative activity provided by inoculation was effective in reducing DM losses. Fermentation products in the control silage are typical of heterofermentative processes, due to a lower lactic acid concentration compared to acetic acid. On the other hand, a homolactic fermentation predominated in inoculated silages. Inoculation also modified the fermentation process through the silo layers. Control and inoculated silages showed higher propionic acid levels in the bottom layer. However, higher butyric acid content was only observed in the control treatment in the bottom layer. This result may indicate that clostridium, which is the main butyric acid producer, benefited from the higher moisture in this layer to grow when the additive was not applied (Table 1).

Table 1. Density (kg DM/m^3), dry matter losses (%) and chemical composition (% DM) in wheat silages with or without homolactic inoculant, according to the silo layer.

	Silage[1]		Silo layer[2]				*P*-value[3]		
Item[4]	C	I	TL	ML	BL	SEM	S	L	S×L
DM (% as fed)	40.2	40.9	42.5^a	41.8^a	37.5^b	0.92	0.23	<0.01	0.11
Density	181	184	138^b	205^a	192^a	35.3	0.12	<0.01	0.37
DML	13.1^A	10.4^B	14.9^a	9.93^b	11.5^b	1.26	0.04	<0.01	0.30
NDF	52.5	53.3	53.7	53.6	51.4	2.98	0.34	0.08	0.12
ADF	32.9	32.3	32.6	32.8	32.4	1.65	0.37	0.85	0.94
CP	9.79^A	8.94^B	9.00	9.23	9.56	0.36	<0.01	0.32	0.18
EE	2.44^A	1.96^B	2.24	2.14	2.22	0.18	<0.01	0.42	0.06
Ash	3.86^A	3.45^B	3.78^a	3.22^b	3.96^a	0.19	<0.01	<0.01	<0.01
IVDMD	62.0	62.4	61.4^b	63.5^a	62.8^{ab}	1.54	0.37	<0.01	0.20

[1] C: wheat silage without inoculants (control); I: wheat silage with inoculant. Means followed by different upper case letters differ within 'Silage' (P<0.05).

[2] TL: top layer; ML: middle layer; BL: bottom layer. Means followed by different lower case letters differ within 'Silo layer' (P<0.05).

[3] Effects of silage (S); layer (L) and interaction (S×L).

[4] DM = dry matter; DML = dry matter losses; NDF = neutral detergent fibre; ADF = acid detergent fibre; CP = crude protein; EE = ether extract; IVDMD = *in vitro* DM digestibility.

Table 2. Fermentation products (% DM) of wheat silage without or with homolactic inoculant.

	Silages[1]		Silo layer[2]				*P*-value[3]		
Item	**C**	**I**	**TL**	**ML**	**BL**	**SEM**	**S**	**L**	**S×L**
pH	4.08^{A}	4.02^{B}	4.09	4.06	4.00	0.03	0.03	0.19	0.43
Lactic acid	1.77	1.87	1.82	1.79	1.85	0.32	0.48	0.94	0.94
Acetic acid	1.91^{A}	0.39^{B}	1.12^{b}	1.22^{a}	1.12^{b}	0.45	<0.01	<0.01	<0.01
Propionic acid	0.53^{A}	0.04^{B}	0.25^{b}	0.18^{b}	0.43^{a}	0.14	<0.01	<0.01	<0.01
Butyric acid	0.57^{A}	0.22^{B}	0.31^{b}	0.38^{b}	0.49^{a}	0.12	<0.01	<0.01	<0.01
Ethanol	0.78	0.81	0.82	0.78	0.78	0.11	0.63	0.79	0.98
$N\text{-}NH_3/NT$	10.3	9.88	10.1	9.94	10.2	0.35	0.45	0.91	0.86

[1] C: wheat silage without inoculant (control); I: wheat silage with inoculant. Means followed by different upper case letters differ within 'Silage' ($P<0.05$).

[2] TL: top layer; ML: middle layer; BL: bottom layer. Means followed by different lower case letters differ within 'Silo layer' ($P<0.05$).

[3] Effects of silage (S); layer (L) and interaction (S×L).

Conclusions

The homolactic inoculant reduced the DM loss during anaerobic storage of wheat silage and affected the fermentation pattern in all layers of large-scale bunker silos.

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Effect of excreta patches on biomass productivity and grazing selectivity in low-input pastures

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Abstract

Grazing animals have a great influence on nutrient cycling and productivity of pastures, especially through the deposition of urine and dung. Animal species differ in pattern of nutrient return and grazing response to excreta. A two-factorial experiment on rotationally grazed pastures was performed, using cattle and sheep as grazer species and grass swards of different botanical composition, either grass-dominated or diverse. Urine, dung and control patches were marked *in situ* in every plot in spring and autumn of 2014. In the following rotation one half of each marked patch was harvested before the next grazing rotation started ('biomass productivity') and the other half after that grazing rotation ended ('forage residue'). The biomass productivity was increased at the urine patches within the next grazing rotation. We also found significantly more biomass remaining at the dung patches following the next grazing rotation, showing an effect of excreta patches on forage residue. Contrary to our initial expectation, the effect of excreta patches on biomass productivity and animal response were neither animal- nor sward-specific.

Keywords: grazing, excreta, biomass productivity, nutrient cycling, low-input pastures

Introduction

The nutrient export from managed pastures in form of animal tissue and other products is very small. Therefore the majority of mineral nutrients ingested by the animals are returned in their excreta (Whitehead, 2000). The productivity of the pasture can be influenced by the spatial concentration of nutrients in dung and urine patches (White-Leech *et al.*, 2013). Apart from the influence on the biomass productivity, excreta patches, especially dung patches, can have an effect on the selectivity of grazing animals (Hirata *et al.*, 1987) and consequently modify the nutrient utilization. To investigate possible interactions between the excreta patch, plant and animal species, a two-factorial experiment with cattle and sheep, which grazed on grass-dominated and diverse pastures, was conducted. Two contrasting observation periods were chosen, with grazing following excreta deposition either after a few weeks or after winter. The effect of excreta patches on plant biomass productivity and animal response were tested. We hypothesized that these values depend on animal species and sward type.

Materials and methods

In a two-factorial experiment, located in the Solling uplands 40 km northwest of Goettingen, with 12 paddocks of a size of 0.5 ha, arranged in 3 blocks, grazed rotationally 3 times per year, the experimental factors animal species (dry Simmental suckler cows and Blackface ewe sheep) and sward type (grass-dominated and diverse sward type) were realized. The initial diversity of the sward was manipulated by the use of herbicide against dicots. The stocking density was 6.27±0.51 for cattle and 4.24±0.29 LU ha^{-1} for sheep (mean ± standard deviation, LU = livestock unit of 500 kg).

Excreta patches were marked *in situ* in 2014, during rotations one (14/05-13/06/2014) and three (29/09-27/10/2014). Following excreta deposition in rotation one, paddocks were grazed again in rotation two (17/06-11/07). After excreta deposition in rotation three, the paddocks were grazed in rotation one of 2015 (11/05-09/06/2015). In each paddock three urine patches were identified by direct animal observation and marked at the end of the respective stocking period. According to the composition of vegetation at the urine patches, dung and control patches were chosen. Around every patch a fence was built. It had an area of 1×1 m for cattle and 0.5×0.5 m for sheep. These patches were fenced in a way that grazing within the fenced areas was possible but the contamination by excrements was prevented.

Immediately before the next stocking period one half of each patch was harvested at stubble height and dry matter yield was determined. The other half was harvested after the stocking period ended, leading to the following target variables: Biomass productivity following different excreta observation periods (cut before animal grazing), animal grazing (difference between the two cuts) and the forage residue (cut following animal grazing).

The data for both observation periods (2014 and 2015) were analysed collectively. The influence of the patch types, observation periods, sward types and animal species on the target variables were examined as fixed factors with mixed models. Block, plot and triplet (urine, dung and control) were used as nested random effects in the models. Models were simplified using Akaike Information Criterion (AIC, Akaike, 1973). The statistical analysis was performed using the program R Statistics (R Core Team, 2015) and the packages nlme (Pinheiro *et al.*, 2015) and lsmeans (Russel *et al.*, 2015).

Results and discussion

The statistical analysis has shown a significant effect for the patch type (P=0.0255) on biomass productivity (Figure 1A). Significantly more biomass was harvested at urine (233.8 g m^{-2}) than at control patches (203.8 g m^{-2}), but biomass productivity was not affected by animal species, sward type or observation period. Animal grazing was neither affected by patch type, nor by animal species, only a significant sward-observation period interaction (P=0.0013) was found. The amount of biomass taken up at grass-dominated patches in spring of 2015 was significantly higher than in summer of 2014, which was probably caused by differences in grazing pressure.

Forage residue (Figure 1B) was significantly affected by patch type (P=0.011). We found significantly more biomass remaining at dung (173.4 g m^{-2}) than at control patches (144.9 g m^{-2}). The forage residue was also significantly affected by the sward-observation period interaction (P=0.0005), having less biomass left at grass-dominated swards in spring 2015 than in summer 2014. The forage residue did not differ between the animal species. Due to the return of most ingested mineral nutrients in excreta (Whitehead, 2000), the excreta deposition leads to different grazer-dependent nutrient balances at small scale (Wrage *et al.*, 2012). Even though the patch effect was not animal-specific, the results are in line with previous observations showing a positive effect of cattle urine patches on plant biomass productivity within 15-20 cm away from the affected area, even in fertilized pasture systems (White-Leech *et al.*, 2013).

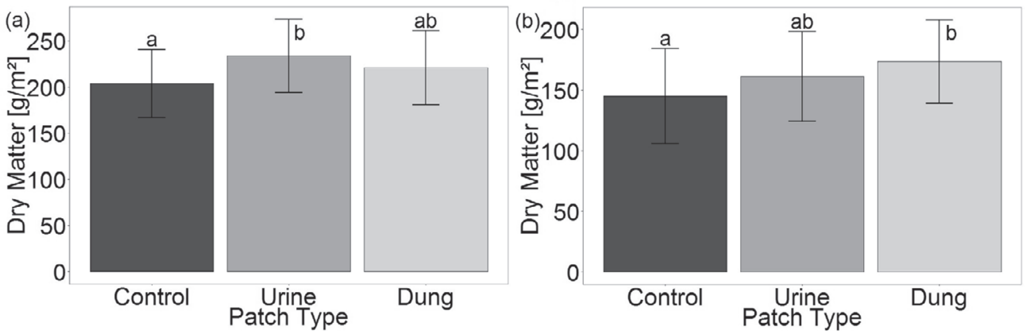

Figure 1. Effect of different patch types on (A) biomass productivity and (B) forage residue, mean values per patch type. Error bars indicate standard error of the dry matter means. Significant differences (P<0.05) between mean values are labelled with different letters.

Grazing was not affected by the patch types, but it has been found for cattle to reject dung patches for 2-18 months (Haynes and Williams, 1993). The animals left significantly more biomass at dung than at control patches, which confirms previous observations (Hirata *et al.*, 1987).

Conclusions

The deposition of excreta significantly affected the plant biomass productivity and forage residue. Only urine patches had a positive effect on plant biomass productivity. Dung patches increased the forage residue. The animal species, sward type and excreta observation period did not modify the excreta effects.

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Variation of digestibility in Italian ryegrass (*Lolium multiflorum* Lam. var. *italicum* Beck)

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Abstract

Among the NW-European fodder grass species Italian ryegrass has the highest yield potential when mown in short-term leys of up to 2 years. However, Italian ryegrass is less digestible in the years after the seeding year than perennial ryegrass because of the presence of stems in almost all cuts. We compared digestibility components of vernalized perennial and Italian ryegrass. Italian ryegrass has a slightly lower content of water soluble carbohydrates (WSC) and its cell wall digestibility (NDFD) is much lower, especially in the summer cuts. In order to evaluate the possibility for improvement of the digestibility of Italian ryegrass by breeding we analysed the variation of digestibility of plot trials sown with varieties and half-sib families of Italian ryegrass. The variation of the NDFD among vernalized families of Italian ryegrass is similar to the variation of the organic matter digestibility (OMD) but much lower than the variation of the content of WSC. The OMD is better correlated to NDFD than to WSC. The variation in NDFD is higher in summer cuts than in spring and autumn cuts. There is no correlation between NDFD and dry matter yield or crude protein content. We may conclude that there is scope for the improvement of the digestibility of Italian ryegrass by breeding for higher cell wall digestibility of the herbage in summer cuts.

Keywords: *Lolium multiflorum,* cell wall digestibility, variation

Introduction

Italian ryegrass is the highest yielding NW-European fodder grass species when mown in short-term leys of up to 2 years. Modern varieties are not heading in the sowing year, resulting in very palatable and digestible herbage. After vernalisation the digestibility of Italian ryegrass is decreasing strongly because of the presence of stems especially from the second cut onwards. The organic matter digestibility (OMD) is mainly determined by the content of water soluble carbohydrates (WSC) and by the cell wall digestibility (NDFD). In these studies we compared these two digestibility components of perennial and Italian ryegrass and studied their variation in Italian ryegrass.

Materials and methods

We compared digestibility components of diploid and tetraploid early heading perennial and Italian ryegrass in a strip plot experiment in three replicates with two levels of nitrogen (N) application rate (350 and 230 kg N ha^{-1} $year^{-1}$) and 2 varieties for each ploidy level of the 2 species. The trial was sown in September 2006 in Merelbeke (Belgium). In 2007 and 2008 the grass plots (size: 8.1 m^2) were cut five times a year. The cultivars were: 'Rebecca' and 'Indiana' for the diploid and 'Merlinda' and 'Lacerta' for the tetraploid perennial ryegrass, 'Bellem' and 'Davinci' for the diploid and 'Gemini' and 'Barmega' for the tetraploid Italian ryegrass. Data were analysed by a factorial ANOVA (Statistica 12.0, Statsoft Inc., Tulsa, OK, USA) with factors year, N-application level and variety.

For the study of the variation in Italian ryegrass, 2 trials were sown in 2012 (D12, T12) and two in 2013 (D13, T13) in Merelbeke (Belgium) with accessions (families and populations) of either diploid or tetraploid Italian ryegrass in 3 replicates. Average heading date of all these accessions is between 11 and 15 May. The trials were mown 5 times in the year after sowing (in 2013 and 2014 resp.), the first being taken in the middle of April, the second in the end of May, the third in the middle of July, the fourth in the end of August and the fifth in the middle of October. The trials received 300 kg of nitrogen fertilizer per ha per year.

At each cut, we determined dry matter yield (DMY) and dry matter content (DMc). A sample of about 300 g fresh weight was taken per plot and dried at 70 °C. The dried samples of the replicates were mixed and ground. We estimated by near infrared spectrometry the WSC content, the OMD, the crude protein (CP) content and the NDFD.

Results and discussion

There was no significant interaction between N-fertilization level and ryegrass variety for all the measured parameters. On average over the 2 N-treatments and 2 years after the sowing year Italian ryegrass is yielding 24% more herbage dry matter than perennial ryegrass (Table 1) with a more than 5%-units lower dry matter digestibility (DMD). The lower digestibility is partly due to a lower WSC content (1.4%-units) but mainly to a much lower cell wall digestibility (7%-units). The lower cell wall digestibility is due to the presence of stems. Van Parijs *et al.* (2014) found that stems of perennial ryegrass had a more than 10%-units lower NDFD than leaves.

The variation of the NDFD among families of Italian ryegrass is similar to the variation of the OMD but much lower than the variation of the content of WSC (Table 2). The cuts in July and August (cut

Table 1. Yield, digestibility and water soluble carbohydrates content of perennial and Italian ryegrass cultivars (average of 2 harvest years, 5 cuts a year and 2 N rates).[1,2]

Ryegrass species	Ploidy	Cultivar	DMY (×1000 kg/ha)	DMD (% /DM)	WSC (%/DM)	NDFD (%/NDF)
perennial	di	Indiana	12.7 cd	67.0 ab	18.4 ab	71.1 b
	di	Rebecca	12.0 d	65.5 b	17.0 bc	69.3 c
	tetra	Lacerta	13.3 bcd	68.6 ab	19.1 a	72.7 ab
	tetra	Merlinda	12.7 cd	67.9 ab	17.9 abc	73.0 a
Italian	di	Bellem	14.9 abcd	62.2 c	16.2 c	64.3 e
	di	Davinci	16.7 a	61.9 c	17.2 abc	65.2 de
	tetra	Barmega	15.8 ab	63.2 c	16.6 bc	66.5 d
	tetra	Gemini	15.3 abc	62.2 c	16.5 bc	66.0 de

[1] Different letters within a column denote significant differences (Duncan's range test, $P<0.05$)

[2] DMY = dry matter yield; DMD = dry matter digestibility; DM= dry matter; WSC = water soluble carbohydrates; NDFD = cell wall digestibility; NDF = neutral detergent fibre.

Table 2. Annual arithmetic means and coefficients of variation of NDFD, WSC content and OMD (%) of accessions in 4 Italian ryegrass trials (n = number of accessions per trial).[1]

trial	n	NDFD		WSC		OMD	
		mean	cv%	mean	cv%	mean	cv%
D12	18	66.7	1.5	20.9	3.8	74.1	1.8
T12	20	69.2	1.2	22.4	4.1	76.3	1.0
D13	32	65.4	1.8	19.3	7.8	73.4	2.3
T13	36	68.4	2.7	22.2	6.7	77.6	2.5

[1] OMD = organic matter digestibility; WSC = water soluble carbohydrates; NDFD = cell wall digestibility.

3 and 4) have the lowest NDFD and the highest variation in NDFD among accessions (Table 3). The annual mean NDFD is best correlated with these cuts 3 and 4 (r=0.68 *** and 0.74*** resp.). The NDFD of the families is very significantly positively correlated with the OMD but almost never significantly correlated with DMY, CP, WSC and DM content (Table 4).

Table 3. Mean cell wall digestibility (%) and variation at each cut in 4 Italian ryegrass trials.

trial	cut1		cut2		cut3		cut4		cut5	
	mean	cv%	mean	cv%	mean	cv%	mean	cv%	mean	cv%
D12	64.0	2.4	73.0	1.4	61.5	4.1	68.3	2.1	–	–
T12	74.8	2.3	68.5	2.3	64.8	2.6	66.5	2.0	71.2	1.7
D13	76.8	3.0	67.6	3.0	57.5	4.3	61.4	3.1	63.5	2.9
T13	76.8	2.4	68.6	2.5	62.1	4.8	65.2	4.7	69.4	3.0
mean	73.1	2.5	69.4	2.3	61.5	4.0	65.3	3.0	68.0	2.5

Table 4. Correlation between NDFD and DMY, WSC content, CP content, OMD and DMc of accessions in 4 Italian ryegrass trials.[1,2]

trial	DMY		CP		WSC		OMD		DMc	
D12	-0.40	ns	0.48	*	0.43	ns	0.92	***	0.18	ns
T12	0.08	ns	0.10	ns	0.07	ns	0.77	***	-0.21	ns
D13	0.04	ns	0.04	ns	0.33	ns	0.79	***	-0.25	ns
T13	0.01	ns	0.15	ns	0.32	ns	0.88	***	-0.41	*
mean	-0.07		0.19		0.29		0.84		-0.17	

[1] *, **, *** significance of Pearson correlation coefficients at respectively $P<0.05$, $P<0.01$ and $P<0.001$; ns: not significant.

[2] DMY = dry matter yield; CP = crude protein; DMc = dry matter content; WSC = water soluble carbohydrates; NDFD = cell wall digestibility; OMD = organic matter digestibility.

Conclusions

The results show that there is scope for the improvement of the digestibility of Italian ryegrass by breeding for higher cell wall digestibility of the herbage in summer. This might simply be achieved by breeding for less stems in summer.

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Inclusion of Arachis pintoi in different levels on diets based on dwarf elephant grass

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Abstract

The aim of this study was to evaluate the effect of different levels of peanut hay inclusion (PE, *Arachis pintoi* cv. Amarillo) on the nutritional value of diets based on dwarf elephant grass (DEG, *Pennisetum purpureum* Shum, cv. Kurumi) hay. Peanut hay was included in the proportions of 0.0, 0.33, 0.66 and 1.00 of total diet dry matter (DM). Eight Texel × Suffolk crossbred wethers provided with duodenal cannula were assigned to the treatments in a replicated 4×4 Latin square. The digestible organic matter (OM) intake increased in diets containing PE hay compared with DEG hay alone, but did not differ between treatments containing PE. Daily N retention as well as duodenal flow of total N, non-ammonia N and microbial N increased in mixed diets compared with DEG alone, whereas PE hay alone showed the highest values for these variables. The inclusion of 330g kg^{-1} DM of PE in DEG based diets is sufficient to improve the digestible OM intake, N retention and can allow N urinary losses to be reduced.

Keywords: *Arachis pintoi*, digestibility, N retention, *Pennisetum purpureum*, voluntary intake

Introduction

The inclusion of forage legumes in ruminant production systems usually improve diet nutritional quality and reduce the need for nitrogen fertilization (Phelan *et al.*, 2015). At moderate levels of legume inclusion (i.e. 300 to 500 g kg^{-1} of total dry matter (DM)), it was observed that *Arachis pintoi* (PE) associated with dwarf elephant grass (DEG; *Pennisetum purpureum* Shum, cv. Kurumi) increased digestible organic matter (OM) intake when compared to the pure grass (Schnaider *et al.*, 2014). However, the nutritional response to higher levels of *Arachis* spp. inclusion was not clearly established. The aim of this study was to evaluate the nutritional response by sheep to increased proportions of PE inclusion in a DEG based diet.

Materials and methods

Treatments consisted of DEG hay offered alone or mixed with PE hay which was included at the proportions of 0.0, 0.33, 0.66 or 1.00 of total diet DM. Eight Suffolk × Texel wethers (average 41±4 kg live weight), fitted with duodenal cannulas and housed in metabolism cages, were used in an experiment with a replicated 4×4 Latin square design. The DEG pasture was fertilized with 50 kg ha^{-1} of N as ammonium nitrate and harvested at 40 cm height from the soil after 35 days of regrowth. The PE hay was produced from plants cut 5 cm height from the soil and no N fertilizer was applied. Hays were chopped in a knife mill with no sieves to 5 to 10 cm particles length, and were offered twice a day (08:00 h and 16:00 h) in amounts to have refusals equivalent to 200 g kg^{-1} of the forage ingested in the previous day. Each experimental period was conducted over 16 d, with a 10 d adaptation and a 6 d measurement period. Feed offered and refused, as well as faeces, were weighed daily and sub-sampled from days 11 to 16 of each experimental period. Urine was collected daily during the measurement period. On day 16, six duodenal samples (50 ml) were collected at 4 h intervals over a 24 h period. All samples were pooled by animal-period for analysis. The duodenal flow of DM (g day^{-1}) was estimated based on acid detergent fibre (ADF) concentration in duodenal digesta and faeces (Kozloski *et al.*, 2014). Data was submitted to variance analysis using the PROC MIXED of SAS using a model that included the random effects of animal and periods, and the fixed effect of legume inclusion levels. When the treatment effect was significant (P<0.05), linear, quadratic and cubic effects of legume inclusion level were tested by using polynomial orthogonal contrasts.

Results and discussion

The nutritional value and chemical composition of experimental forages were presented in Table 1. The OM intake linearly increased (P<0.001), whereas total apparent OM digestibility linearly decreased (P<0.001) increasing the dietary proportion of PE (Table 2). The digestible OM intake increased in diets containing PE hay compared with DEG hay alone, but did not differ between treatments containing PE from 330 until 1000 g kg^{-1} of DM (quadratic effect: P<0.05). The N intake and N urinary excretion

Table 1. Nutritional value and chemical composition (g kg^{-1} DM) of experimental forages.[1]

Parameter	DEG	PE
Dry matter (g kg^{-1} fresh)	800	798
Organic matter	874	905
Crude protein	98	167
Neutral detergent fibre	668	572
Acid detergent fibre	405	467
Metabolisable energy (Mcal kg^{-1} DM)	1.82	1.71

[1] DM = dry matter; DEG = dwarf elephant grass; PE = *Arachis pintoi*.

Table 2. Intake, digestibility, duodenal flow, urinary excretion N, faecal excretion, retention N, ruminal undegradable protein and ruminal degradable protein in sheep fed dwarf elephant grass hay alone or with inclusion of *Arachis pintoi* hay.

	Proportion of *Arachis pintoi* hay					*P*-value			
	0	0.33	0.66	1.00	SEM	ANOVA	Linear	Quadratic	Cubic
Intake (g/day)									
DM	1,063	1,184	1,234	1,279	29.1	<0.001	<0.001	0.198	0.610
OM	922	1,046	1,100	1,155	26.2	<0.001	<0.001	0.195	0.544
Digestible OM	560	625	628	616	16.0	0.001	0.031	0.025	0.520
Nitrogen	20.2	25.3	30.7	37.7	0.76	<0.001	<0.001	0.199	0.750
Total apparent OM digestibility	0.62	0.60	0.57	0.52	0.005	<0.001	<0.001	0.133	0.558
Ruminal apparent OM digestibility	0.53	0.54	0.51	0.43	0.005	<0.001	<0.001	<0.001	0.316
Duodenal flow (g day^{-1})									
Total N	19.0	22.7	23.5	34.5	0.89	<0.001	<0.001	0.012	0.017
Non-ammonia N	18.8	22.4	23.1	33.5	0.83	<0.001	<0.001	0.012	0.015
Microbial N	12.1	14.6	14.9	21.5	0.56	<0.001	<0.001	0.001	0.015
Urinary excretion N (g day^{-1})	7.6	9.8	12.3	13.4	0.15	<0.001	<0.001	0.257	0.471
Faecal excretion (g day^{-1})									
DM	395	502	559	646	12.5	<0.001	<0.001	0.433	0.169
N	8.7	11.6	14.2	17.4	0.27	<0.001	<0.001	0.622	0.462
Retention N:									
g day^{-1}	3.5	3.9	3.8	7.0	0.41	<0.001	<0.001	0.004	0.057
g g^{-1} N intake	0.15	0.15	0.12	0.18	0.011	0.023	0.446	0.009	0.070
RUP (%)	35.6	25.6	24.8	26.6	2.1	0.031	0.034	0.051	0.483
RDP (%)	65.5	74.3	75.1	73.4	2.1	0.031	0.034	0.051	0.483

[1] DM = dry matter; OM = organic matter; RUP = ruminal undegradable protein; RDP = ruminal degradable protein; SEM = standard error of the mean.

increased linearly ($P<0.01$) with increasing dietary proportion of PE. The N retention, duodenal flow of total, non-ammonia and microbial N increased in mixed diets compared with DEG alone, with PE hay alone showing the highest values for this variable (cubic effect: $P<0.06$).

The digestible OM intake increased approximately 12% due the inclusion of 330 g kg^{-1} of DM of PE in the DEG based diet, which is similar to results of Schnaider *et al.* (2014), who found a 14% increase in the digestible OM intake by including 300 g kg^{-1} DM of PE hay in a DEG based diet offered to sheep. However, at higher levels of legume inclusion no additional nutritional advantages were observed, since increased OM intake was offset by decreased total apparent OM digestibility. These results can be, at least partially, explained because the cell walls in legume forages are less resistant to chewing and microbial degradation than the cell walls of grasses. Thus, by increasing the PE proportion in diet probably increased the rates of particle comminution and passage from rumen (Beever and Thorp, 1996), decreasing total apparent OM digestibility.

The lack of difference on N retention observed between treatments 330 and 660 g kg^{-1} DM of PE inclusion, as well as the highest N retention by animals receiving PE pure hay were coherent with the results of intestinal flow of non-ammonia N. The flow of non-ammonia N is primarily a result of microbial protein synthesis, since the inclusion of peanut decreased the proportion of ruminal undegradable protein as compared to the pure grass diet. On the other hand, the peanut inclusion clearly increased the N intake and, despite of have also increased urinary N excretion, it also improved the N retention in peanut pure treatment compared with offering only DEG or mixed diets. This result could be explained, at least partially, due to the absence of N fertilization on peanut pastures. It is well known that N fertilization has a direct effect decreasing the proportion of plant N present as true protein (Peyraud and Astigarraga, 1998) and rumen microbial growth is stimulated by amino acids and peptides compared with ammonia as a N source (Argyle and Baldwin, 1989).

Conclusions

The inclusion of PE at a ratio of up to 330 g kg^{-1} of DM in mixed diets with DEG is sufficient to maximize the digestible OM intake and N retention, and can allow N urinary losses to be reduced. Higher inclusion levels of PE may be accepted without negative effect on digestible OM intake.

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Intake and performance of ewes and lambs fed grass-clover silage treated with chemical additives

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Abstract

The aim was to investigate the effect of grass-clover silage treated with chemical additives on intake and performance of pregnant and lactating ewes and their lambs. The forage (34% dry matter (DM)) was treated with GrasAAT Plus (formic, propionic, benzoic acids and their salts, 3 L/T) and Kofasil Ultra K (nitrite, hexamine, sorbate, benzoate, propionate, 2 L/T) during baling, which were compared with baled silage without additive. The silage contained 11.4 MJ metabolisable energy kg^{-1} DM and was fermented well. Data were analysed in a completely randomized design using seven twin bearing ewes per treatment during late pregnancy and lactation. Lamb data were analysed until slaughter. Silages were supplemented with concentrate. There were no treatment effects on intake and body condition of the ewes. Lambs born from ewes fed treated silages had higher birth weights (5.8 vs 5.2 kg, P=0.01) and live weight gains from birth to weaning (439 vs 409 g day^{-1}, P=0.03) than lambs from ewes fed untreated silage. Weaned lambs fed treated silages had higher DM intakes (P=0.02). Lamb performance from birth to weaning was improved indicating improved nutrient utilisation by ewes fed silage treated with chemical additives.

Keywords: additive, sheep, silage

Introduction

Grass silage intake and performance by ewes in late pregnancy and lactation has been shown to be affected by maturity stage of the grass at harvest and by physiological state of the ewe (Nadeau *et al.*, 2015a). Grass silage fermentation products, such as organic acids and alcohols can affect intake by ruminants (Gerlach *et al.*, 2013; Krizsan and Randby, 2007). Use of additives improves grass silage fermentation characteristics, which can result in improved nutrient utilisation and performance by ruminants (Nadeau *et al.*, 2015b). The objective of this experiment was to determine the effect of additive-treated grass-clover silage on intake and performance of pregnant and lactating ewes and their lambs.

Materials and methods

The sward (75% *Phleum pratense* L. and *Lolium multiflorum* L. × *Festuca pratensis* L./25% *Trifolium pratense* L. of dry matter (DM)) was mowed on June 1, 2011, prewilted to 34% DM and ensiled in round bales. The forage was treated with GrasAAT Plus (formic, propionic, benzoic acids and their salts, 3 L/T, Addcon Nordic AS) and Kofasil Ultra K (nitrite, hexamine, sorbate, benzoate, propionate, 2 L/T, Addcon Europe GmbH) during baling, which were compared with baled silage without additive. Chemical composition of the silages was analysed by conventional methods and metabolisable energy (ME) was calculated from *in vitro* rumen organic matter digestibility. Twenty-one twin-bearing Swedish Finewool × Dorset ewes, mated with a Dorset ram, were randomly allocated to the three silage treatments with seven ewes per treatment. The ewes were fed 0.5 kg concentrate in addition to silage *ad libitum*. The lambs were fed concentrate and silage *ad libitum* until weaning at 56 (standard deviation (SD)=5.0) d of age and thereafter 0.8 kg concentrate and silage *ad libitum* until slaughter at 44-45 kg. The initial mean body weight (BW) and body condition of the ewes was 96.5 (SD=9.17) and 3.6 (SD=0.36), respectively. Feed intake was registered daily. Ewes and lambs were weighed once a week and ewes were scored for body condition once a week. Spot samples of urine from the ewes during lactation were taken morning and afternoon on one day for analysis of allantoin and uric acid. Daily renal excretion of the purine derivatives allantoin and uric acid was calculated according to Chen and Ørskov (2003). Lamb carcasses were classified according to the EUROPE scale, where 5-15 is the approved interval for conformation and 4-8 is the approved interval for fatness. Ewe data were analysed statistically in a completely randomized design, using PROC MIXED of SAS (ver. 9.3). Period (late pregnancy and lactation) and treatment were fixed effects and ewe nested within treatment was a random effect. Lamb data were analysed similarly with treatment and sex as fixed effects and ewe nested within treatment as a random effect. Results on effects of silage treatment on intake and performance of ewes and lambs will be presented in this paper. Significance is declared at $P<0.05$, tendency to significance at $0.05<P<0.10$ and non-significance at $P>0.10$. LSMEANS are separated by Tukey's adjustment (LSMEANS with different superscripts differ) and by using the contrast 'additive vs untreated silage'.

Results and discussion

The silage contained 140 g crude protein, 477 g neutral detergent fibre (NDF) and 11.4 MJ ME kg^{-1} DM. All silages were fermented well (pH: 4.4-4.5, lactic acid: 51-73 g, acetic acid: 9-11 g, sugar: 93-129 g kg^{-1} DM; NH_3-N: 68-89 g kg^{-1} total N) with no differences between treatments, except for ethanol, which was 5 g kg^{-1} DM for additive-treated silage and 13 g kg^{-1} DM for untreated silage. There were no treatment effects on silage DM intake (2.17 and 3.25 kg d^{-1} in late pregnancy and lactation, respectively), silage NDF intake (11.6 and 16.6 g kg^{-1} BW in late pregnancy and lactation, respectively), total crude protein (CP) intake (423 and 575 g d^{-1} in late pregnancy and lactation) and total ME intake (30.7 and 43.3 MJ d^{-1} in late pregnancy and lactation, respectively). Furthermore, body condition of the ewes did not differ between treatments (3.5 and 2.9 in late pregnancy and lactation, respectively). Renal excretion of the purine derivatives from the lactating ewes did not differ between silage treatments (17.3, 18.3 and 19.6 g d^{-1} for untreated, GrasAAT Plus and Kofasil Ultra K, respectively), which indicates that microbial protein synthesis was similar between ewes fed the different silage treatments (Chen and Ørskov, 2003).

The live weight (LW) of the lambs at birth was, on average, 0.6 kg higher and LW gain from birth to weaning was 30 g higher for the lambs born from ewes fed the additive-treated silages compared to lambs born from ewes fed the untreated silage (Table 1). Daily intakes of DM, NDF, CP and ME after weaning were higher for lambs fed the additive-treated silages than for lambs fed the untreated silage. Carcass traits of the lambs were similar between treatments, except for the fatness, which was scored higher for lambs fed the additive-treated silages (Table 1). The scores were all within the approved range according to the EUROP classification scheme.

Conclusions

Increased live weight at birth and increased live weight gain from birth to weaning of the lambs indicate improved nutrient utilisation by the ewes fed silage treated with the chemical additives. The increased intake of the additive-treated silages by the weaned lambs did, however, not result in increased live weight gain from weaning to slaughter.

Table 1. Intake, live weight, live weight gain and carcass traits in lambs fed untreated or additive-treated grass-clover silage.[1,2]

	Untreated	GrasAAT Plus	Kofasil Ultra K	SEM	*P*-value	
					Treatment	Additive vs Untreated
Intake weaning – slaughter						
Silage DM, kg d^{-1}	0.55^{b}	0.72^{a}	$0.64^{a,b}$	0.040	0.03	0.02
Total DM, kg d^{-1}	1.26^{b}	1.43^{a}	$1.35^{a,b}$	0.040	0.03	0.02
Silage NDF, g kg^{-1} BW	6.13^{b}	8.21^{a}	$7.40^{a,b}$	0.046	0.02	<0.01
Total NDF, g kg^{-1} BW	11.5^{b}	13.5^{a}	$12.7^{a,b}$	0.044	0.02	0.01
Total CP, g d^{-1}	243^{b}	267^{a}	$251^{a,b}$	5.8	0.02	0.03
Total ME[3], MJ d^{-1}	15.3^{b}	17.5^{a}	$16.5^{a,b}$	0.47	0.01	<0.01
Live weight, kg						
At birth	5.2^{b}	$5.6^{a,b}$	6.0^{a}	0.18	0.02	0.01
At weaning	29.1	29.4	30.1	0.64	ns	ns
At slaughter	44.4	45.0	44.5	0.63	ns	ns
Live weight gain, g d-1						
Birth to weaning	$409^{(b)}$	$436^{(a,b)}$	$442^{(a)}$	10.8	0.09	0.03
Weaning to slaughter	397	414	371	16.5	ns	ns
Birth to slaughter	403	429	410	11.8	ns	ns
Carcass traits						
Carcass weight, kg	19.7	19.5	20.0	0.34	ns	ns
Dressing, %	44.5	43.3	44.9	0.60	ns	ns
Conformation	9.4	9.6	10.2	0.36	ns	ns
Fatness	6.7^{b}	$7.2^{a,b}$	7.8^{a}	0.28	0.04	0.03
Age at slaughter, d	93.4	87.9	89.2	2.97	ns	ns

[1] DM = dry matter; NDF = neutral detergent fibre; BW = body weight; CP = crude protein; ME = metabolisable energy; SEM = standard error of the mean; ns = not significant.
[2] Values within a row with the same superscript letter do not differ significantly.

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Towards improved potassium fertiliser recommendation for silage maize in the Netherlands

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Abstract

Optimal potassium (K) fertilisation stimulates maize production. In 2015 a two year K fertiliser trial on land sown with maize was started in order to update the 40 year old recommendation. Farm field experiments were performed on 20 locations on sandy, clay and peat soils with varying K availability and buffering capacity. There were 6 treatments on each site: 0, 40, 80, 160 kg K_2O ha^{-1} applied as mineral fertiliser and cattle slurry (CS) application (40-45 m^3 $ha^{-1,}$) with an additional 0 or 80 kg K_2O ha^{-1} applied as mineral fertiliser. In parallel, three detailed experiments with different K fertiliser levels (0-320 K_2O ha^{-1}) were performed; two on sandy and one on clay soil. Potassium was applied as mineral fertiliser and CS. Broadcast application was also compared with band application of K_2O. The N level in the trials was 200-250 kg N ha^{-1} to ensure no N deficiency. Fertilisation with K_2O resulted mostly in non-significant dry matter yield increases. However, there was a strong relationship between K uptake and K fertiliser rate and the potassium and magnesium soil status (determined in 0.01M $CaCl_2$). The experiments will be continued in 2016.

Keywords: potassium, fertilisation, soil fertility, CEC, silage maize, DM yield

Introduction

Optimal maize growth requires an adequate supply of potassium (K). A desk study gave indications that the K fertilization recommendation could be improved (Van Schooten *et al.*, 2012). Firstly, the Dutch K fertiliser recommendation for silage maize is mainly derived from other crops and grain maize trials of about 50 years ago. Secondly, silage maize yield has increased over the last decades. Thirdly, there is also potential for improvement of the soil K-status determination. The current one is based on a K-index (based on 0.1 M HCl soil extraction and soil organic matter content). Measuring available K in 0.01 M $CaCl_2$ soil extracts in combination with the cation exchange capacity (CEC) seems a promising method (Van Rotterdam, 2010).

Cattle slurry (CS) is the main K source for silage maize. Due to the manure application regulations in the Netherlands, the application of CS is restricted to about 35-40 m^3 ha^{-1}, equalling 200-250 kg K_2O ha^{-1}. This indicates that farmers have to apply additional mineral K_2O fertiliser in case of a low soil status. This might not be necessary, since the German recommendation is lower than the Dutch. Taking all this into account, a two year K fertiliser experiment on multiple locations was started in 2015. This paper summarizes the first results.

Materials and methods

The 2015 experiment consisted of two sub-experiments: farm field trials on 20 locations, and detailed trials at 3 locations. The farm field trials were performed on clay, peat and sand soils with 2 sites per location varying in K-availability and buffering capacity (Table 1). There were 6 treatments on each site: 0, 40, 80 and 160 kg K_2O ha^{-1} (as potassium sulphate) and CS application (40-45 m^3 ha^{-1}) plus 0 and 80 kg K_2O ha^{-1}. In total there were 240 experimental plots. Together with the maize sowing 30 kg N ha^{-1}

Table 1. The soil analysis results of the 40 sites used in the farm field trial and of the 3 sites in the detailed trials.[1]

	Clay (g kg^{-1})	Silt (g kg^{-1})	Sand (g kg^{-1})	OM (kg N ha^{-1})	CEC (mmol+ kg^{-1})	pH[2]	K[2] (mg kg^{-1})	Mg[2] (mg kg^{-1})	P[2] (mg kg^{-1})	PAL[2] (mg P_2O_5 100 g^{-1})
40 sites										
Min	490	440	900	89	365	7.3	168	347	14.9	154
Mean	123	199	624	44	124	5.7	81	125	4.1	65
Max	10	50	100	25	31	4.3	23	35	0,6	34
Detailed trials										
Sand 1				3.5		5.3	44			71
Clay				5.9		6.3	33			51
Sand 2				5.1		5.1	75			75

[1] OM = organic matter; CEC = cation exchange capacity; PAL = P_2O_5 extracted with ammonium lactate.

[2] Extraction with 0.01 M $CaCl_2$.

was band-applied. To achieve an N level of 200 kg N ha^{-1} at every site, N fertiliser (CAN) was applied by broadcasting.

The detailed trials were setup on sandy (2 sites) and clay soil (1 site). The treatments were:

- no CS and 0, 40, 80, 160 or 320 kg K_2O ha^{-1}, broadcast and band application;
- 20 m^3 CS ha^{-1} broadcast and 0, 40, 80 or 160 kg K_2O ha^{-1}, broadcast and band application;
- 20 m^3 CS ha^{-1} row application and 0, 40, 80 kg K_2O ha^{-1}, broadcast and band application.

The setup was a randomized block design with 4 replicates. Together with the maize sowing, 25 kg N and 7 kg P_2O_5 ha^{-1} was band applied. Fertiliser N (CAN) was applied to achieve an N level of 200 kg N ha^{-1}. Plots were harvested between mid-September and November. Herbage yield and mineral composition were determined. The maize yield data were statistically analysed using REML (Harville, 1987) with location as random factor.

Results

In the farm field trials yield varied from 10.6 to 25.2 t dry matter (DM) ha^{-1} between locations. The average yield was 18.4 t DM ha^{-1} and average K content was 11.4 g K kg^{-1} DM, varying from 5.3 to 19.2 g K kg^{-1} DM. The average K, N and P uptake was 210 kg K, 225 kg N and 37.1 kg P ha^{-1}. The DM yield response to K_2O fertilisation was small. Only at a low soil K-status (Table 2) was a significant DM yield response found. Maize yield was positively related to soil K-status (P<0.05). There was a positive relationship between K_2O application and K uptake (Table 2). This was most apparent at a low soil K-status. Statistical analysis indicated a strong relationship between K uptake and K fertiliser rate (P<0.05) and soil potassium (P<0.01) and magnesium status (P<0.01).

In the detailed trial we observed a clear crop response of fresh yield to K_2O fertilisation. However, based on DM only a weak yield response was found (Figure 1), despite the low soil status. The DM content of fertilised treatments was higher than that of unfertilised treatments. Figure 1 shows that maximum yield was achieved with less than 100 kg K_2O ha^{-1}. Average yield at the 3 sites was respectively 20.1, 14.9 (sandy soils) and 16.6 (clay soil) tonnes DM ha^{-1}. The K uptake showed a response to about 200 kg K_2O ha^{-1} (data not shown), comparable with the farm field trials. The experiments showed that band application of K_2O was not more effective than broadcast application. This was the case for both DM yield and for K uptake.

Table 2. Mean dry matter (DM) yield and K uptake for the different treatments of the farm field trials (n=40) grouped to a K status which is respectively <50, <80 and <100 mg kg^{-1}.

	DM yield t ha^{-1}			K-uptake kg K_2O ha^{-1}		
	Mean (n=7)	Mean (n=19)	Mean (n=27)	Mean (n=7)	Mean (n=19)	Mean (n=27)
K soil (mg kg^{-1})	<50	<80	<100	<50	<80	<100
0 kg K_2O ha^{-1}	17.3a	18.1a	18.3a	142a	186a	197a
40 kg K_2O ha^{-1}	18.6ab	18.8a	18.5a	158ab	193ab	201ab
80 kg K_2O ha^{-1}	18.7ab	18.6a	18.8a	168ab	202bc	209bc
160 kg K_2O ha^{-1}	18.4ab	18.2a	18.7a	173b	206c	215c
CS+ 0 kg K_2O ha^{-1}	18.9ab	17.8a	18.1a	175b	212c	221cd
CS + 80k g K_2O ha^{-1}	20.0b	19.0a	18.9a	207c	225d	226d
sed	0.78	0.56	0.47	15.4	8.5	6.8
Sign.[1]	*			***	***	***

[1] Significance* $P<0.05$, ** $P<0.01$ and *** $P<0.001$.

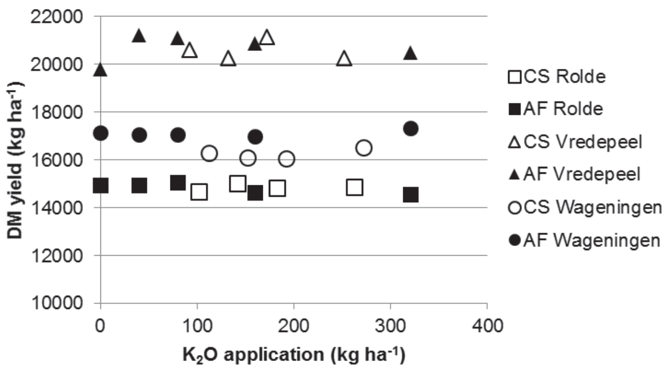

Figure 1. The effect potassium fertilisation on dry matter (DM) yield of maize at three locations with 20 m^3 ha^{-1} cattle slurry application (CS) and with only mineral fertiliser (AF).

Discussion

In trials 50 years ago, Prummel (1966) found a strong reponse only at a low soil status (K-HCl <80 mg K_2O kg^{-1}). In recent trials the K response in maize is low. A yield response in corn of about 10% was observed only at a very low soil status (K-HCl <100 mg K_2O kg^{-1}) (Kovacevic *et al.*, 2009). A 3 year trial in Canada showed a yield effect only in one year when no K was applied (Subedi *et al.,* 2009). Besides soil status, crop K content can be an indicator of adequate K supply. Leigh and Jones (1984) suggest a critical crop content of 13 g kg^{-1} DM. About 80% of the data in our trials showed lower values than 13 and about 45% even lower than 10 g kg^{-1} DM. In combination with the low yield response to K fertilisation this suggests that the critical K content is much lower 13 g kg^{-1} DM. The trials will be continued in 2016.

Conclusions

The indication so far is that K fertilisation recommendation levels for silage maize can be reduced in the Netherlands. The soil analysis results indicate that a single parameter, K measured in 0.01 M $CaCl_2$ will do. The experiments will be continued in 2016.

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Impact of inoculation with homofermentative bacteria on the aerobic stability of wheat silage stored in farm-scale bunker-silos

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Abstract

The objective was to examine effects of inoculating whole crop wheat with homolactic bacteria (*Lactobacillus plantarum* plus *Pediococcus acidilactici* at 1×10^5 cfu g^{-1}) on the profile of fermentation end-products and aerobic stability across different layers of large-scale bunker silos. Wheat crop was harvested at soft dough stage (~40% of dry matter (DM)), packed in two bunker silos (30 t $silo^{-1}$) and the treatments were settled as 2 (control vs inoculant) × 3 (bottom vs middle vs top layer) factorial arrangement, with twelve replicates per layer (completely randomized design). Lactic, acetic, propionic and butyric acids, and ethanol were measured by chromatographic analysis. An aerobic stability test was performed for 7 d. Silage concentrations of ethanol and lactic acid were not affected by treatments, although there was an interaction between inoculant and silo layer for the concentrations of volatile fatty acids (VFA). Inoculated silages had lower concentrations of acetic, propionic and butyric acids in all layers compared with the control silage. In untreated silage, the middle layer had more acetic acid, whereas butyric and propionic acid contents were greater in the bottom layer. The lower content of VFA in silages treated with homolactic bacteria led to worse aerobic stability. In conclusion, the homolactic inoculant decreased the formation of VFA and aerobic stability of wheat silages stored in bunker silos.

Keywords: aerobic deterioration, bunker silo, *Lactobacillus plantarum*, *Triticum aestivum* L.

Introduction

The primary goal of making silage is to maximize the preservation of nutrients. In order to improve the fermentation process, silage additives based on homofermentative bacteria have been used to increase the recovery of nutrients and, in some cases, animal performance (Kung *et al.*, 2003). However, in many instances, homolactic inoculants lead to silages that are less stable when exposed to air (Muck and Kung, 1997), probably due to a lower content of antifungal end-products, such as acetic acid. Additionally, most studies reporting the effects of silage inoculants were carried out in lab scale silos. Thus, the objective of this work was to examine effects of inoculating whole crop wheat with homolactic bacteria on the fermentation end-products and aerobic stability in different layers of large-scale bunker silos.

Materials and methods

Wheat cultivar BRS Umbu was sown in 3 hectares, using a row spacing of 0.17 m and a stand density of 220 seeds m^{-2}, in Guarapuava-PR, Brazil (25°24'S 51°28'W). At soft dough stage, the whole crop wheat (~40% dry matter (DM)) was mechanically harvested (Pecus 9004 PRN 1200) and packed in two 30-t bunker silos (3.5 m width, 11 m length and 1.2 m height), using a tractor weighting approximately 6 tons. Silos were filled simultaneously (every second load for each silo) in one working day and sealed with polyethylene film (black-on-white, 200 μm). At ensiling, 2 treatments (control vs inoculant) × 3 silo layers (bottom = 0 to 40 cm vs middle = 40 to 80 cm vs top = 80 to 120 cm layer) were factorially arranged, with 12 replicates per layer (one bunker per silage type). The inoculant contained *Lactobacillus plantarum* MA 18/5U and *Pediococcus acidilactici* MA 18/5M and was applied at a theoretical rate of 1×10^5 cfu $g^{-1}$of forage. At silo filling, twelve tracer bags (12×50 cm, nylon 85 μm pore size) containing 1.5 kg of pre-ensiling forage were allocated throughout each silo layer and used for sampling. Silos were unloaded simultaneously, with a feedout rate of 14 cm d^{-1} (on average). Lactic, acetic, propionic and butyric acids, and ethanol were measured by chromatographic analysis. An aerobic stability test was performed for 7 d. Approximately 1.2 kg of silage was loosely added to plastic buckets and exposed to air at 20±1.5 °C. Aerobic stability was defined as the time (hours) until silage temperature reached 2 °C above ambient temperature (Moran *et al.*, 1996). Mean daily silage temperatures above ambient temperature were accumulated over 7 d of aerobic exposure and used as a marker of aerobic deterioration (O'Kiely, 2009). Data was analysed as a completely randomized design using the GLM procedure of SAS (SAS Institute Inc., Cary, NC, USA), including the effects of inoculant, silo layer and their interaction. Treatment means were compared by Tukey test (α=0.01).

Results and discussion

As expected, silage from the top (85 h) was less (P<0.01) stable than silage from middle and bottom silages (average of 93 h). Moreover, the homofermentative inoculant markedly decreased the aerobic stability (42 vs 129 h) of wheat silages stored in bunker silos. Similar results have been reported for wheat silage treated with homolactic inoculant in lab scale silos (Weinberg *et al.*, 2010). The concentrations of ethanol (average of 0.79% of DM) and lactic acid (average of 1.82% of DM) were neither affected by inoculant nor by silo layer (P>0.48). However, there was an interaction (P<0.01) between inoculant and silo layer for the concentrations of volatile fatty acids (VFA). Inoculated silages had lower concentrations of acetic, propionic and butyric acids in all silo layers (P<0.01). In untreated silage, the middle layer had more acetic acid, whereas butyric and propionic acid contents were greater in the bottom. Therefore, treating wheat with homolactic bacteria intensified the aerobic deterioration, due to the lower content of VFA (Figure 1).

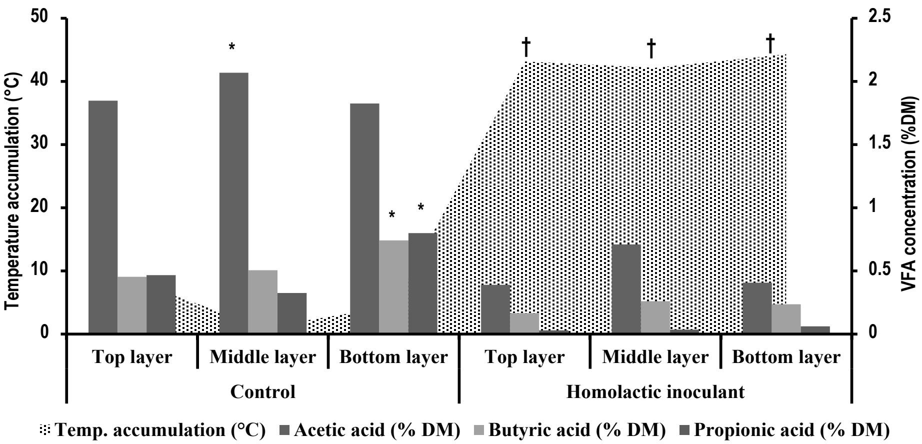

Figure 1. Aerobic deterioration and volatile fatty acids (VFA) concentrations in wheat silage stored in bunker silos. † P<0.01 for inoculant effect. * P<0.01 for silo layer effect within the control treatment. DM = dry matter.

Conclusions

The homolactic inoculant decreased the formation of VFA and worsened the aerobic stability of wheat silages stored in bunker silos.

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Modelling the surface of a paddock affected by urine and faeces deposition during grazing by dairy cows

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Abstract

In grazed grassland, urine and faeces, returned by grazing animals and added to the nitrogen (N) from fertilizer, augments the N fluxes and also the risk of N losses (leaching, ammonia volatilization). Urine and faeces deposition by the grazing animal result in large applications of N to a small area within the paddock. Many agronomic models do not take into account this aspect of grazing systems. As the area affected by urine and faeces at each deposition is reported to be 2 m^2, the Moorepark Grass Growth model (MGGm) describes the N fluxes in a grid basis at a 2 m^2 level. The MGGm assumes that within any given grazing event the localisation of the urine and faeces deposition will be random with overlapping being a possibility. The objective of this paper is to compare the consequences of a uniform deposition over the paddock versus localize at 2 m^2 individual animal depositions, in four different grazing simulations. At 2 m^2 level, the urine and dung deposition induces a huge variation in term of mineral N available (maximal difference of 335 kg of N), grass growth (maximal difference of 5,052 kg of DM) and N leached (maximal difference of 134 kg of N leached). At the paddock level, the uniform simulation has a slightly higher net grass growth and N uptake compared to the 2 m^2 simulation. Finally, within a simulation, the average N leached evaluated between November and April of year 2 are identical between the uniform and a 2 m^2 localized applications.

Keywords: grass growth, urine patches, N leached, model

Introduction

Nitrogen (N) fertilizer is highly effective at increasing grass growth and hence farm productivity, but it also contributes to nitrogen (N) leaching to groundwater (Delaby *et al.*, 1997). On a grass grazed paddock, urine and dung depositions from grazing animals increase the N added through fertilization which increase the N availability for grass growth and the risk of N losses (leaching, volatilization) on a small portion of a paddock area. Traditionally soil models simulate urine patches uniformly across the whole paddock. The objective of this study is to use the Moorepark Grass Growth model (MGGm – Ruelle and Delaby, 2016) to show the heterogeneity of growth and N leached in a grazed paddock due to grazing animal depositions and to evaluate the consequences of a uniform or a 2 m^2 localized approach at the paddock level.

Materials and methods

The MGGm is a dynamic model developed in C++ describing the grass growth and the nitrogen flux of a paddock at a 2 m^2 level. The core model functions are described in Ruelle and Delaby (2016). The main inputs of the model are the type of soil (in terms of clay, sand and organic matter content), grassland management (date of grazing or cutting event, number of animals, post grazing cutting height ...), fertilisation management (date and quantity of N applied), and the daily weather (in terms of temperature, rain and radiation). In this model, the faeces and urine depositions are considered to affect only a 2 m^2 area of the paddock. Therefore, the paddock is represented as a grid of 2 m^2 having individual mineral N, organic N, grass biomass and grass N content. The localisation of each deposition on that grid is random with overlaps. The model has been tested for different weather conditions (Table 1). Two

Table 1. Description of the two annual weather data applied in the simulations.

Location Year	Annual average temperature (°C)	Monthly rainfall (mm)												Annual total rainfall (mm)
		J	F	M	A	M	J	J	A	S	O	N	D	
France Normandie	10.2	61	32	136	68	64	12	35	72	42	80	102	41	743
Ireland Co. Cork	8.7	107	39	88	59	38	53	143	23	102	83	98	37	869

different N mineral applications have been simulated (zero or 150 kg N per ha (France) /200 kg of N per ha (Ireland) per year, in six or height applications respectively). The size of the simulated paddock is 1 ha and there are a total of seven (France) or eight (Ireland) grazing events. The initial N mineral status of the soil was 80 kg per ha and the N organic level was at 14,400 kg N per ha (6% of OM content). In the simulations, the number of animals for each grazing event was calculated depending on the pre grazing height (calculated by the model) based on a daily intake of the animal of 16 kg of dry matter (DM).

To test the impact and the importance of modelling at a 2 m^2 level, the simulations have been run either with the normal feature of the model or either with the urine and faeces deposition considered as uniform applications across the paddock. The simulations were completed across 2 years with the same weather and management conditions allowing the quantification of the impact of the first grazing year on the leaching that occurred during the winter and spring of the second year. The N leaching results presented are the corresponding N leaching between 1 November of year one to 30 April of year two (called the winter period). All the other results concern only the first grazing year of the simulation. To highlight the heterogeneity within a paddock for the 2 m^2 simulation, results in the Table 2 are presented on the form of minimal (Min), maximal (Max) and standard deviation (SD). The Min and Max represent the 2 m^2 with the lower or higher value of the variable considered, the SD represents the standard deviation of those values within a paddock. Due to the variable considered (net growth, N mineral and N leached), the Min for each variable corresponds to an area of 2 m^2 which received no urine or faeces deposition.

Results and discussion

Table 2 describes the variability in terms of grass growth across the year and the mineral N content in the soil on the first of November and N leached during the winter period within a paddock when simulated at the 2 m^2 scale. According to the number of grazing days realised, the surface of a paddock affected by a urine deposition was of 52, 63, 65 and 78% for the French study with 0 N, French study with 150 N, Irish study with 0 N and Irish with 200 N, respectively. When there was an increase in mineral N fertilisation,

Table 2. Internal heterogeneity of the paddock in grazing condition. The maximal (Max), minimal (Min) and standard deviation (SD) of the net grass growth, N mineral and the total N leached, of the 2 m^2 within a paddock.

		Net growth (kg of DM)		N min at 1 Nov.		N leached	
N (France / Ireland)		0	150/200	0	150/200	0	150/200
France Normandie	Min	6,575	8,544	86	119	34	45
	Max	10,909	12,080	201	454	67	116
	SD	829	732	15	39	4	9
Ireland Co.Cork	Min	8,226	11,564	91	125	54	72
	Max	13,278	16,573	194	411	102	206
	SD	901	902	13	35	7	18

there was an increase in grass growth, N mineral soil and N leaching. Accumulation of urine patches induce a huge increase in grass growth between 3,536 and 5,052 kg of DM per ha in the simulations. Similarly, the N leaching differs between the different 2 m^2 areas with a maximal difference in the Irish simulation at 200 N (increase of 134 kg N leached). The lowest maximal difference in N leached is in the French simulation with no fertilisation with a difference of 33 kg N leached. At the paddock level, when comparing the localized with the uniform simulation (Table 3), the grass growth is slightly higher in the uniform simulation (average of 118 kg of DM per ha higher over the year) which led to a higher number of grazing days. This difference is slightly higher in the simulation with mineral N fertilization than in the one without. When looking at the N leached and the N mineral at the start of the winter, both simulations are strictly identical.

Table 3. Comparison between the uniform and the localized simulation on the total net grass growth, the total N uptake, the number of grazing day, the N in the urine of the animal the N mineral in the soil and the total N leached of the year on the weather and the N mineral fertilisation (N)

		Net growth (kg dry matter)		**N uptake (kg N/ year)**		**Number of grazing days**		**N urine (kg per year per ha affected)**		**N min at 1 Nov.**		**N leached**	
N (France / Ireland)		**0**	**150/200**	**0**	**150/200**	**0**	**150/200**	**0**	**150/200**	**0**	**150/200**	**0**	**150/200**
France Normandie	Uniform	7,412	9,453	141	243	381	507	51	98	97	150	37	52
	Localized	7,336	9,350	139	237	376	501	98[1]	153[2]	97	151	37	52
Ireland Co. Cork	Uniform	9,243	12,972	173	336	531	770	56	135	103	157	61	90
	Localized	9,146	12,777	169	327	524	758	88[3]	171[4]	103	159	61	91

[1] On 52% of the paddock.
[2] On 63% of the paddock.
[3] On 65% of the paddock.
[4] On 78% of the paddock.

Conclusions

Integrated at the paddock level, the impact of uniform or localized urine and dung patches has been very small under those grazing conditions. However, it would be interesting to investigate the impact in a system where the cows are supplemented and stay on a paddock with a relatively small amount of grass in the diet.

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The role of fungal diseases in the accumulation of dead tissue in timothy and meadow fescue swards

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Abstract

Under Nordic climate conditions the proportion of dead tissue in silage leys can be substantial in the second and third cuts and thus decrease the nutritive value of the yield. The role of plant diseases in this process is insufficiently known. Our aim was to determine the effects of fungal diseases on accumulation of dead tissue and the nutritive value in timothy (*Phleum pratense* L.) and meadow fescue (*Festuca pratensis* Huds.) swards. A field experiment was conducted at Maaninka, Finland during 2011-2014. The factors were: timing of the second harvest and the use of chemical control of fungal diseases. After the first cut in June half of the plots were sprayed with a mixture of fungicides Amistar and Zenit. The second cuts were taken 4, 6 or 8 weeks after the first cut. The proportion of dead tissue was 0.6-11.2% in timothy and 2.2-27.0% in meadow fescue, increasing with longer cutting interval. In meadow fescue the fungicide treatment decreased the proportion of dead tissue and simultaneously increased digestibility in one year out of three. The effect was strongest at the latest cutting time. In timothy the effects were minor.

Keywords: dead tissue, fungal diseases, fungicide, meadow fescue, timothy

Introduction

To achieve high yield and quality, it is important to understand the mechanisms affecting herbage mass production and development of nutritive quality of grass forage. Under Nordic climate conditions, accumulation of dead herbage tissue is typical, especially in regrowths (Kuoppala *et al.*, 2008; Virkajärvi *et al.*, 2012) when day length and incoming solar radiation are decreasing. A substantial proportion of dead tissue can reduce the nutritive value of the entire herbage yield. The senescence rate in a grass sward is mainly determined by the ontological life span of leaves (Virkajärvi *et al.*, 2012), competition for light (Parsons, 1988) and plant diseases (Plumb, 1988). The role of plant diseases in the production of senesced tissue of grasses is largely unknown. In this study the aim was to determine the importance of fungal diseases for yield, quality and accumulation of dead material in the herbage yield of the two commonly cultivated grass species in northern Europe, timothy and meadow fescue.

Materials and methods

Field experiments were conducted at Maaninka (63°08' N, 27°19' E), Finland, between 2011 and 2014. The study was carried out as a split plot experiment with four replicates. The main plot was the timing of the second harvest (Time (T); 4, 6 and 8 weeks after the first cut) and the subplot was the use of chemical control of grass diseases (Fungicide (F); yes or no). There were separate experiments with timothy and meadow fescue swards. The first cut was taken in June at the same time for all plots, and the date of second cut depended on the T-treatment. The plots were fertilized according to the Finnish recommendations. F-treatments were sprayed after the first cut with a mixture of two fungicides (Amistar (Azoxystrobin) 0.3-0.4 l ha^{-1}, Zenit (Propiconazole + Fenpropidin) 0.3-0.4 l ha^{-1} in water at 300 l ha^{-1}). Before each harvest the proportion of the sward affected by plant diseases was estimated visually and samples were taken to identify the cause. Dry matter (DM) yield, digestibility (D-value; *in vitro* cellulose; Huhtanen *et al.*, 2006) and proportion of senesced tissue were determined for each cut. Statistical analyses were performed using Mixed-procedure of SAS 9.3. Replicates and years were considered to be random effects

Table 1. The effect of harvest timing (T) and chemical control (F) of fungal diseases on the proportions of the sward affected by diseases and proportions of dead tissue of timothy and meadow fescue swards in the second harvest in years 2011-2013.

		Proportion of sward with diseases, %						Proportion of dead tissue, %					
T	F	Timothy			Meadow fescue			Timothy			Meadow fescue		
		2011	2012	2013	2011	2012	2013	2011	2012	2013	2011	2012	2013
4 week	no	1	1	1	3	5	2	1.2	0.6	1.3	4.4	4.1	4.5
4 week	yes	1	1	1	3	4	3	7.2	0.9	1.0	5.3	2.2	5.4
6 week	no	2	2	4	6	9	8	9.4	7.9	2.8	14.0	12.8	12.7
6 week	yes	2	2	4	6	8	7	9.8	3.6	2.0	13.9	10.3	11.3
8 week	no	5	8	6	13	24	11	11.2	7.2	4.8	17.7	27.0	23.3
8 week	yes	4	9	4	10	11	7	8.7	7.2	3.0	14.8	14.6	20.6
SEM[1]		0.3	0.9	0.4	0.5	0.8	0.5	1.53	0.89	0.35	1.25	1.00	1.22
Signif.[2]	T	***	***	***	***	***	***	**	***	***	***	***	***
	F			***	*	***	**		o	**		***	
	T × F			***	*	***	***	*	*			***	

[1] SEM = standard error of mean.
[2] Significance *** P<0.001; ** P<0.01; * P <0.05; o P<0.10.

while T, F and their interaction were considered fixed effects. Regression analysis was used to explore the effect of temperature sum and occurrence of diseases on D-value of forage.

Results and discussion

The most abundant pathogenic fungi in meadow fescue was *Drechslera dictyoides* and in timothy *Drechslera phlei*. In addition, *Mastigosporium* sp. was observed in 2013 in timothy. The proportion of diseased plants was higher in meadow fescue than in timothy swards (Table 1). In both species longer cutting intervals resulted in an increase of plant diseases. The influence of F-treatment was significant in meadow fescue each year and significance increased with the growing time. In timothy the effect was statistically significant in 2013. The proportion of dead tissue was 0.6-11.2% in timothy and 2.2-27.0% in meadow fescue and increased with longer cutting interval (Table 1). In meadow fescue the F-treatment decreased the proportion of dead tissue significantly in 2012. The effect was strongest in the latest harvest. No statistical differences were observed in 2011 or 2013. In timothy the effects were minor despite being statistically significant in 2013.

In this study the F-treatment increased yield only in the latest harvest of meadow fescue in 2011 (Table 2). In contrast, in timothy the F-treatment slightly decreased DM yield (average 200 kg DM ha^{-1}) in 2011 and 2013. Decreased yield could be explained an injurious effect caused by the F-treatment in 2011 and therefore the fungicide dose was reduced from 0.4 to 0.3 l ha^{-1} for the last two years. The F-treatment increased D-value in meadow fescue in 2012. The difference between the treatments was 10, 12 and 32 g kg^{-1} DM for 4-, 6- and 8-week cutting intervals, respectively. In timothy the effect was opposite in 2011, which can be explained by fungicidal injury to the grass. D-value of dead tissue samples varied between 544 and 690 g kg^{-1} DM.

This study was purely a scientific investigation, as fungicides are not accepted for use in grass leys in Finland. Their effect here was minor but there are some cases where small increases in DM yield have been reported (Alamikkotervo, 1996). Linear regression analysis revealed that temperature sum explained 76

Table 2 The effect of harvest timing (T) and chemical control of fungal diseases (F) for dry matter (DM) yield and digestibility (D)-value of timothy and meadow fescue swards in the second harvest in years 2011-2013.

		DM yield, kg DM ha^{-1}						D-value, g kg^{-1} DM					
T	F	Timothy			Meadow fescue			Timothy			Meadow fescue		
		2011	2012	2013	2011	2012	2013	2011	2012	2013	2011	2012	2013
4 week	no	2,520	1,190	1,170	2,580	1,880	1,740	699	738	758	718	739	776
4 week	yes	2,240	1,130	1,100	2,480	1,750	1,710	689	737	761	717	749	782
6 week	no	4,710	3,360	3,390	4,260	3,470	3,040	682	687	716	719	706	767
6 week	yes	4,520	3,210	3,150	4,210	3,530	3,070	675	690	714	718	718	765
8 week	no	5,730	4,950	5,400	4,560	4,240	3,970	665	644	664	691	657	732
8 week	yes	5,190	4,970	5,050	4,710	4,280	3,950	661	655	661	700	689	742
SEM[1]		195	58	85	115	81	134	3.8	3.9	4.3	5.8	6.0	5.2
Signif[2].	T	***	***	***	***	***	***	***	***	***	*	***	***
	F	*		**				*				**	o
	T × F				*							o	

[1] SEM = standard error of mean.

[2] Significance *** $P<0.001$; ** $P<0.01$; * $P<0.05$; o $P<0.10$.

and 30%, and fungal diseases 48 and 51%, of the variation in D-value in timothy and meadow fescue, respectively. It should be noted that there was a strong positive correlation between temperature sum and occurrence of fungal diseases.

Conclusions

The amount of dead tissue in the second harvest was higher in meadow fescue than in timothy. The proportion of dead tissue in the yield increased when time between cuts was extended. The significance of fungal diseases in yield formation was minor, but fungal diseases increased the amount of dead tissue in the yield of meadow fescue resulting in decreased digestibility.

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Early lactation pasture allowance and duration: the effect on bodyweight and body condition score in cows

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Abstract

Feed deficits can occur in grazing systems due to low spring grass growth. The objective of this experiment was to investigate if different pasture allowances (PA) offered to early lactation grazing dairy cows for varying time durations influenced bodyweight (BW) and body condition score (BCS; 5 point scale). The two-year experiment offered cows one of four PA (60, 80, 100 or 120% of intake capacity), for either 2 or 6-weeks. Once the 2- and 6-week time durations had elapsed, the cows were offered 100% PA. At the end of the first 2 weeks, there was a linear increase in BW with increasing PA (441, 449, 456 and 464 kg, respectively), but there was no effect of PA on BCS (2.99). There was no effect of the interaction between PA and duration on BW at the end of week 6; BW increased linearly with increasing PA (443, 453, 456 and 466 kg; 60, 80, 100 and 120%, respectively), but, there was no effect of PA or duration on BCS at the end of 6-weeks (2.91). After 10-weeks there were no differences in BW or BCS. However, these results cannot be examined in isolation; other variables such as milk production and fertility need to be considered in order to obtain a complete picture of the residual effects of altering PA in early lactation.

Keywords: pasture allowance, early lactation, bodyweight, body condition score, dairy cow

Introduction

Increased pasture utilisation can reduce the feed costs associated with dairy production systems and increase profitability (Shalloo *et al.*, 2004). An increase in grass utilisation is generally achieved with lower post grazing sward heights, but this results in lower pasture allowances being offered to grazing dairy cows (Ganche *et al.*, 2013). Furthermore, feed deficits can lead to lower pasture allowances in grazing systems due to low spring grass growth which can result in the mobilisation of body reserves as cows try to balance the deficit between reduced energy intake and early lactation milk production requirements. The objective of this experiment was to investigate if different pasture allowances (PA) offered to early lactation grazing dairy cows for varying time durations influenced bodyweight (BW) and body condition score (BCS; 5 point scale).

Materials and methods

The study was undertaken over a two year period; from March 25 to November 27, 2014 and March 9 to November 23, 2015 at the Teagasc Moorepark research farm, Fermoy, Co. Cork, Southern Ireland. Each year 96 dairy cows (41 primiparous and 55 multiparous in year one (Y1); 24 primiparous and 72 multiparous in year two (Y2)) were assigned to a randomised complete block design with a 4×2 factorial arrangement of treatments. Cows were completely re-randomised in 2015. Cows were balanced and randomly assigned to one of four PA (60, 80, 100 or 120% of intake capacity; IC) for either 2 or 6 weeks. Once the 2- and 6-week time durations had elapsed, the treatments were offered 100% of IC. No concentrate was offered to cows during the experiment. Intake capacity was calculated using the equation of Faverdin *et al.* (2011) based on age, parity, days in milk, BW, BCS and potential milk yield. Pastures were >80% perennial ryegrass. Fresh pasture areas were offered after each milking during the treatment period and on a 24-hour basis thereafter. Pre- and post-grazing sward heights were measured daily using a rising plate meter. Herbage mass (HM; >3.5 cm) was measured twice weekly. Treatment groups grazed adjacent to each other to ensure similar HM was offered. Pasture allowance (>3.5 cm) for the 60, 80 and 120% treatments were calculated based on the IC of the 100×6 treatment, the mean IC value of all animals in the 100×6 group was used. As HM was similar between treatments daily area allocations were different. An electronic portable weighing scale with the Winweigh software package (Tru-test Limited, Auckland, New Zealand) was used to record BW weekly. Body condition was scored by the same individual throughout the study on a weekly basis using a scale from 1 to 5 (where 1 = emaciated and 5 = extremely fat) with 0.25 increments (Edmondson *et al.*, 1989). Data were analysed using covariate analysis and mixed models in SAS v9.3. Fixed terms for year, parity, breed, PA, duration and the interaction of PA and duration were included, with cow as the random effect. Pre-experimental values were used as covariates.

Results and discussion

The mean PA for the 60, 80, 100 and 120% treatments for the first two weeks were 8.0, 10.7, 13.4 and 16.0 kg dry matter (DM) cow^{-1} day^{-1}, respectively (P<0.001). This resulted in post-grazing heights (PGH) of 2.4, 2.9, 3.5 and 4.1 cm, respectively (P<0.001). The mean PA and PGH during weeks 3-6 were 9.1, 12.1, 15.1, 18.2 kg DM cow^{-1} day^{-1}and 2.5, 3.1, 3.8, 4.3 cm for the 60, 80, 100 and 120% 6-week treatments. PA and PGH for the 2-week treatment, which was grazed as a single herd during weeks 3-6, were 14.8 kg DM ha^{-1} and 3.7 cm, respectively.

There was no interaction between PA and duration on BW or BCS. At the end of the first 2 weeks of the experiment, BW increased linearly with increasing PA (441, 449, 456 and 464 kg, respectively; Figure 1). For each percentage increase in PA cow BW increased by 0.38 kg. At the end of the six-week experimental period, there was no interaction between PA and duration but there was an effect of PA (P<0.001) and duration (P<0.01) on cow BW. Similar to the first two-weeks of the experiment BW increased linearly with increasing PA (443, 453, 456 and 466 kg, respectively). At the end of the six week experimental period the linear increase in BW was the same as at the end of the first two-weeks – for each percentage increase in PA cow BW increased by 0.38 kg. Bodyweight was similar for all PA treatments at the end of the tenth week (464 kg). Kennedy *et al.* (this volume) reported that the 60x6 treatment reduced their yield of milk fat plus protein during this six-week period, indicating that these cows reduced their energy output rather than excessively mobilising body reserves to maintain milk production. Given the

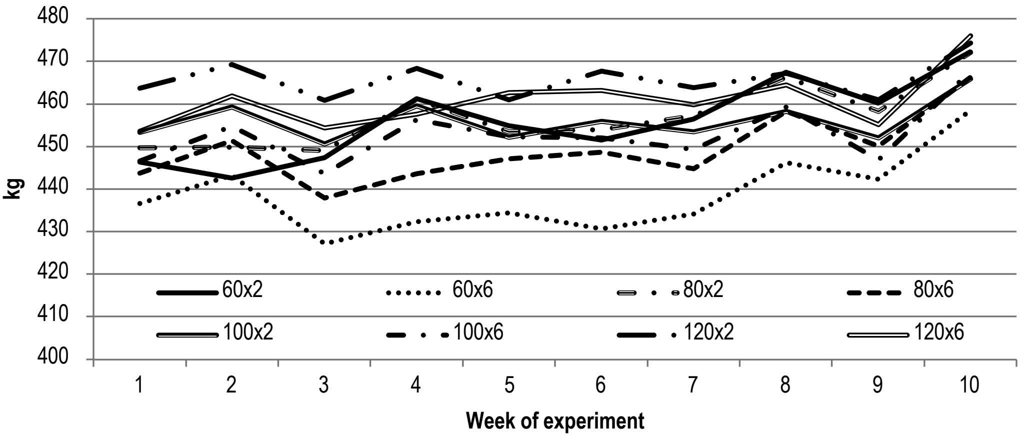

Figure 1. Bodyweight profile of early lactation dairy cows offered 1 of 4 pasture allowances (60, 80, 100 or 120% of intake capacity) for either 2 or 6 weeks.

difference between the treatments (<20 kg) they may have been a result of differences in gut fill. This is further reinforced when BCS was observed as there was no effect of PA or duration on the herd average BCS after either two (2.99) or six (2.91) weeks on treatment.

Conclusions

Any differences in BW observed when different PAs were imposed may have been due to differences in gut fill as no difference in BCS at any of the time points were evident. However, the results of this experiment need to be considered alongside milk production, fertility and behaviour and welfare measurements to draw definite conclusions and to understand the true effect of offering different PA for different durations of time to cows in early lactation.

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The development of yield and digestibility of the third cut of grass silage in Finland

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Abstract

The three-cut strategy for making grass silage is now becoming more common in eastern and northern parts of Finland. The development of yield and digestibility of grass at the third harvest is not well understood. This paper summarizes the results of six 3- and 4-year experiments (2009-2015) to provide information about the characteristics of the third cut. Three experiments were located in North Savo, two in North Ostrobothnia and one in Kainuu. Timothy, meadow fescue or a mixture of these two species was used. The experimental design, the amount of nitrogen fertilization and the timing of harvests varied between experiments. The data contained a total of 77 observation points. The dry matter yield of the third cut varied between 790 and 4,130 kg dry matter (DM) ha^{-1}. The average daily growth rate during the observed period was 35.3 kg DM ha^{-1} d^{-1}. The amount of DM yield was well explained by the date of the second harvest (R^2=0.72) and the growth was affected by temperature sum (R^2=0.54). D-value was always above 680 g kg^{-1} DM and its daily change was -0.7 g kg^{-1} DM d^{-1}.

Keywords: dry matter yield, D-value, grass, harvest timing, third cut

Introduction

The three-cut strategy for grass silage is now becoming more common in eastern and northern parts of Finland. The weather conditions near the end of growing season differ from the conditions of the earlier growing season. Due to rapidly shortening day length, temperature sum accumulates more slowly and the amount of radiation decreases. Soil moisture can vary from excessively dry at the beginning of growth, to excessively wet during the autumn harvest. The development of yield and digestibility of the third harvest is not well understood. This paper summarizes the results of six harvest strategy experiments to provide information on the relationship between weather parameters and characteristics of the third harvest.

Materials and methods

Six field experiments were carried out in 2009-2015, each lasting three or four years. The experimental design, cultivars and the timing of harvests varied between experiments. Three experiments were located in Maaninka (63°08′ N, 27°19′ E), two in Ruukki (64°68′ N, 25°09′ E) and one in Sotkamo (64°11′ N, 28°33′ E). Timothy (*Phleum pratense* L.), meadow fescue (*Festuca pratensis* Huds.) or a mixture of these two species was used. The third harvests were fertilized with 30-55 kg N ha^{-1}, 0-30 kg K ha^{-1} and 0-5 kg P ha^{-1} depending on the experiment. Dry matter (DM) yield, digestibility (D-value), neutral detergent fibre (NDF) and crude protein (CP) were determined as well as leaf stem ratio in part of trials. The data were derived by calculating averages of replicates for each treatment. Most of the nutritive values in the data (70) were analysed by near infrared spectrometry (Valio Ltd.) and seven at Luke, as follows: D-value using *in vitro* cellulose (Huhtanen *et al*, 2006), NDF according Van Soest *et al* (1991) and CP using Kjeldahl-method. Weather data were obtained from the Finnish Meteorological Institute. Regression analyses were performed using the *Reg*-procedure of the SAS 9.3.

Results and discussion

Weather data, DM yields and the corresponding parameters of nutritive values for the third harvest are presented in Table 1. The dates of the second and the third cut and growing times of the third harvest varied depending on experimental designs. Delaying the third cut increased the accumulated temperature and radiation sum only marginally. The DM yield of the third harvest varied a lot (Table 1). D-value was always above 680 g kg^{-1} DM, although the amount of dead material was substantial in some cases. The mean proportion of leaves was 0.79 of living DM. The average D-value of 716 g kg^{-1} DM was higher than the 670 g kg^{-1} DM reported previously for a large third-harvest farm-data set (Salo *et al.,* 2014). Digestibility of the third harvest in the study of Salo *et al.* (2014) was also higher than in the second harvest (664 g kg^{-1} DM). The average NDF content was 494 g kg^{-1} DM, which is lower than typical NDF values in the second harvest (535 g kg^{-1} DM; Salo *et al.*, 2014). The CP content varied a lot but the average was low compared with the average values of second- and third-cut harvests (145 g kg^{-1} DM and 148 g kg^{-1} DM, respectively) as reported by Salo *et al* (2014). This can be explained by the lack of organic fertilizers in experiments compared with the typical management used on farms.

The date of the second cut was the best estimate for DM yield in the third cut (Table 2; R^2=0.72). The effective temperature and radiation sums explained DM yield equally well (R^2=0.54 and R^2=0.55, respectively). In contrast, the date of the third cut was not significant. The changes in NDF were more dependent on the date of the second cut, temperature sum and radiation than the changes in D-value (Table 2). Average daily growth rate during the observed period was 35 kg DM ha^{-1} d^{-1}. The daily change of D-value was -0.7 g kg^{-1} DM d^{-1}. For comparison, Virkajärvi *et al.* (2012) reported that the growth rate of the second harvest near the cutting time was 78 kg DM ha^{-1} d^{-1} in timothy and 83 kg DM ha^{-1} d^{-1} in tall fescue (*Festuca arundinacea* Schreb.). According to Kuoppala (2010) the daily decrease of D-value in the second harvest was 1.4 g kg^{-1} DM d^{-1}. Taking a third harvest is an option that allows increased annual DM yield without a decrease in grass D-value, assuming that growing conditions are favourable (Hyrkäs *et al.*, 2015). The most effective way to increase the DM yield of the third harvest is to cut the second harvest earlier; this would reduce its DM yield but increase the digestibility. Typically, the D-value of the second harvest is considerably lower than the D-value of the third harvest, which

Table 1. Summary of cutting dates, weather data, dry matter yields and parameters of nutritive value in the third cut in 2009-2015 (n=77).

		Min	Mean	Max	SD[1]
Second cut date		11 July	24 July	11 Aug.	
Third cut date		28 Aug.	13 Sep.	11 Oct.	
Growing time (from 2nd cut to 3rd cut)	d	26	51	77	13.2
Temperature	°C	11.2	14.8	17.5	1.42
Precipitation	mm d^{-1}	1.0	2.2	3.1	0.37
Radiation	MJ m^{-2} d^{-1}	8.2	13.4	16.4	1.98
Effective temperature sum[2,3]	°C d	249	488	696	118.1
Precipitation sum[2]	mm	40	109	207	33.6
Radiation sum[2]	MJ m^{-2}	406	668	945	153.4
Dry matter (DM) yield	kg DM ha^{-1}	790	2,600	4,130	836
D-value	g kg^{-1} DM	682	716	758	19.9
Neutral detergent fibre	g kg^{-1} DM	434	494	561	30.6
Crude protein	g kg^{-1} DM	78	125	198	29.1

[1] SD = standard deviation.

[2] From the previous cut onwards.

[3] Base temperature 5 °C.

Table 2. Linear regression equations and coefficients of determination (R^2). Dates of second and third cut are presented as the number of days from the beginning of May.[1]

Y	X		Intercept	Slope	R^2
DM yield (kg DM ha^{-1})	Date of second cut	d	9,485	-81.3	0.72
	Date of third cut	d	2,647	-0.35	0.00
	Number of growing days	d	809.0	35.3	0.31
	Effective temperature sum[2,3 b)]	°C d	70.1	5.18	0.54
	Radiation sum[2]	MJ m^{-2}	-100	4.04	0.55
D-value (g kg^{-1} DM)	Date of second cut	d	594	1.44	0.40
	Effective temperature sum[2,3]	°C d	767	-0.11	0.40
	Radiation sum[2]	MJ m^{-2}	766	-0.08	0.34
NDF (g kg^{-1} DM)	Date of second cut	d	738	-2.88	0.67
	Effective temperature sum[2,3]	°C d	394	0.21	0.63
	Radiation sum[2]	MJ m^{-2}	399	0.14	0.51

[1] DM = dry matter; D-value = digestibility value; NDF = neutral detergent fibre.
[2] From the previous cut onwards.
[3] Base temperature 5 °C

makes this option appealing. Grass growth in September is so low that delaying the third cut has only a minor effect on DM yield. There is also evidence that the chemical feed analysis for the third harvest may overestimate its production value (Sairanen *et al*, 2016). In addition, the low NDF content has to be taken into account when planning animal feeding. In the future, growing seasons are predicted to become longer (Ruosteenoja *et al.*, 2011), which would increase the potential for utilization of autumn growth.

Conclusions

Due to the slower accumulation of temperature sum and radiation sum towards the end of the growing season, together with absence of true stem, changes of quality and the amount of grass yield in the third harvest differ from the first and second harvests. Daily changes of D-value and DM yield are relatively small in the third harvest and delaying the harvest time in September has only a minor effect on the grass yield. The most effective way to increase the DM yield of the third harvest is to cut the second harvest earlier.

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Timing of different non-chemical control strategies of narrow-leaved ragwort (*Senecio inaequidens*) in grassland

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Abstract

Narrow-leaved ragwort (*Senecio inaequidens*) is an invasive alien species and a poisonous weed containing pyrrolizidine alkaloids (PA). It usually spreads in rural environments along communication ways such as railroads and road embankments, but it can occasionally thrive also in dry grassland. As PA cause irreversible liver damage to animals and can also be retrieved in the milk of dairy animals, a control of this weed is advisable to ensure an adequate forage quality. In order to define viable control strategies, the effect of mowing or hand-pulling, carried out at four different phenological stages (before flowering, at flowering, at seed ripening and at the end of the growing season), was investigated in a four-year field experiment in a mountain environment in the Venosta Valley (South Tyrol, Italy). Once a year the density of weed plants, subdivided into four different size classes, was assessed before the earliest treatment. Hand-pulling rapidly decreased the proportion of medium-sized and large-sized plants, while mowing did not apparently change the proportion between size classes. Hand-pulling was twice as effective as mowing, and the highest effectiveness was achieved if the treatment was applied towards the end of the growing season.

Keywords: *Senecio inaequidens*, non-chemical weed control, hand-pulling, mowing

Introduction

Narrow-leaved ragwort (*Senecio inaequidens* L.) is an invasive alien species native of Southern Africa, which was accidentally introduced in Europe at the end of the 19th century and widely spread since then (Heger and Böhmer, 2005). It reproduces mainly generatively and thrives usually in fallow, uncultivated areas, spreading along roadsides and railways (Ernst, 1998), but was occasionally observed in South Tyrol to spread also in extensively managed dry pastures. In such situations, ragwort plants are avoided by grazing animals and, if pasture maintenance is neglected, they can reproduce and spread, leading to deterioration of productivity and forage quality of the pasture. Narrow-leaved ragwort is a poisonous weed containing pyrrolizidine alkaloids (PA), which cause irreversible liver damage to animals and can also be retrieved in the milk of dairy animals (Hoogenboom *et al.*, 2011). Mowing and hand-pulling are current measures to control grassland weeds. Reports about their effectiveness in controlling narrow-leaved ragwort already exists in the literature (EPPO, 2006; Heger and Böhmer, 2005), but there is scarcity of experimental data and of information about the most suitable timing of treatment application, which is a pivotal issue of non-chemical control strategies.

Materials and methods

A field experiment to investigate the efficacy of repeated mowing or hand-pulling to reduce the density of ragwort plants over time was conducted from 2012 to 2015 in a south-exposed, dry pasture with sparse shrubs at Platzerböden (Laces/Latsch, Alto Adige/South Tyrol, Italy; 1,150 m.a.s.l., 4.9 °C yearly mean temperature and 500.4 mm year^{-1} precipitation according to the weather station of Silandro/Schlanders), which exhibited since 2008 increasing abundance of narrow-leaved ragwort. Mowing and hand-pulling were applied at four different phenological stages of the ragwort plants: before flowering (30% floral buds), at flowering (30% flowering capitula), at seed ripening (30% developed achenes) and in late autumn (Table 1). A control treatment was included as well. The experiment was laid out as a two-factorial randomised complete block design with three replicates and a plot size of 36 m^2 (6 6 m). Ragwort plant density was recorded in spring (between 4 and 11 April), assigning the counted plants to four size classes: young plants (1 stem $plant^{-1}$), small plants (2 to 9 stems $plant^{-1}$), medium plants (10 to 30 stems $plant^{-1}$), large plants (more than 30 stems $plant^{-1}$). Treatment efficacy on plant density between the first and the last observation year was calculated with the method of Henderson and Tilton (1955), which also takes in account the changes of plant density in the control plots to assess treatment efficacy. Statistical analysis was performed by ANOVA. Data were checked for normal distribution of residuals and heteroscedasticity. Multiple comparisons were performed by LSD. A *P*-value <0.05 was considered to be significant.

Results and discussion

The results of ANOVA showed that both the control method (P=0.005) and the timing of treatment (P=0.048) had an effect on the efficacy of the treatment in reducing the density of *Senecio* plants, while there was no significant effect of their interaction (P=0.461).

Hand-pulling was about twice as effective as mowing; treating at the end of the growing season was more effective than any other timing of treatment (Figure 1). During the observation time, the density of *Senecio*-plants in the control plots showed a certain fluctuation (1.78 plants m^{-2} in 2012, 2.72 plants m^{-2} in 2013, 2.43 plants m^{-2} in 2014, 1.81 plants m^{-2} in 2015) but not a clear trend, suggesting the

Table 1. Timing of treatment application from 2012 to 2014.

Treatment application	Year		
	2012	2013	2014
Before flowering	22 May	14 June	13 June
At flowering	26 June	8 July	2 July
At seed ripening	18 July	3 August	8 August
Late Autumn	20 November	30 November	30 October

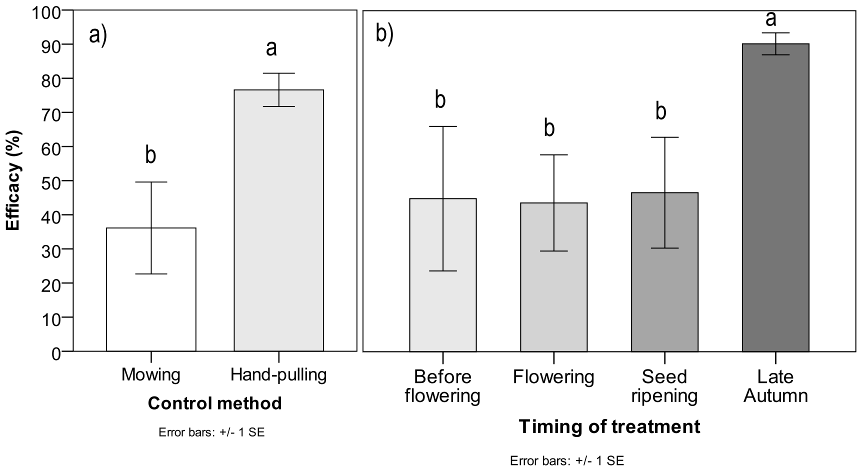

Figure 1. Efficacy of non-chemical control of *Senecio inaequidens* between the first and the last observation year depending on (A) the control method and (B) the timing of treatment. Means without letters in common significantly differ from each other.

occurrence of some mortality and turnover pattern depending on the meteorological conditions. Taking into account the changes over time of the density of plants belonging to different classes (Figure 2), it can be observed that mowing results in the mid-term in a partial survival of the small, medium and large plants, whilst hand-pulling strongly reduced most classes, with a partial survival of the small class. Hand-pulling caused a strong reduction of the plant density already after the first treatment.

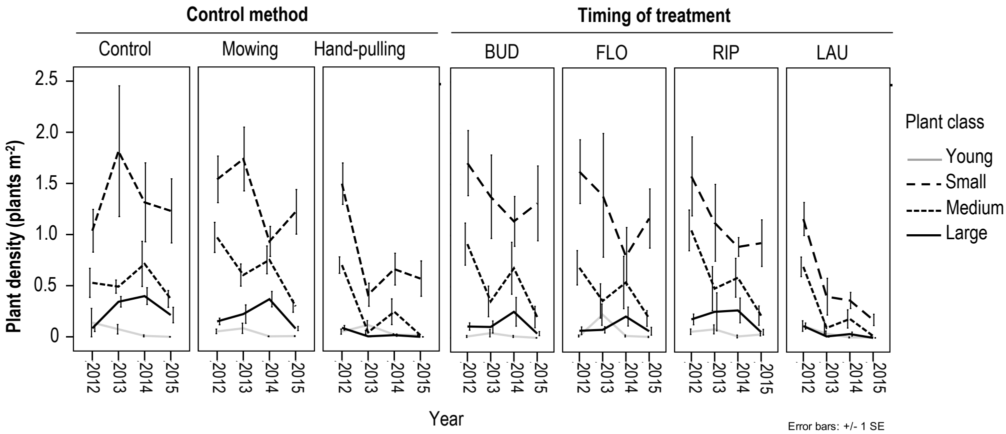

Figure 2. Changes over time in the density of different size classes depending on the control method and on the timing of the treatment. BUD = before flowering, FLO = flowering, RIP = seed ripening, LAU = late autumn.

Conclusions

Hand-pulling was the most effective method to control *Senecio inaequidens*. Late treatments at the end of the growing season seemed to be advisable. However, the amount of plants possibly escaping the counts in spring after a treatment in late autumn, as well as the germination dynamic of seeds, represent an uncertainty factor and deserve further investigation. Because of the considerable time effort associated with hand-pulling, an intervention at an early stage of infestation is of great relevance.

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The relation between stocking rate, supplementary feed and grazing hours on grass intake as assessed by model simulations

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Abstract

The number of grazing cows is decreasing in several countries in Europe. Farmers sometimes have problems finding the right grazing system suitable for their farm, especially in situations with restricted grazing, and therefore make the choice to keep the herd inside. Guidelines will help farmers to choose the right grazing system and the right number of grazing hours adapted to the amount of supplementary feed and the stocking density. To optimise grassland management, farmers need predictions of grass intake under a range of supplementary silage feeding and stocking densities. Based on model calculations with the whole farm model DairyWise, a table with an overview of grass intake was created for different stocking densities and different amounts of supplementary silage feeding in the Netherlands. The model used a mowing percentage of 200% (the total grazing area was mown twice a year for silage) to realise enough aftermath. The supplementary feeding was based on a TMR with 50% silage maize and 50% silage grass. The table is applicable for an average herd (including primiparous and multiparous dairy cows) with an average annual milk production of about 8,500 kg cow^{-1} and gives a quick overview of the grazing possibilities.

Keywords: grass intake, grazing, stocking density, supplementary feeding

Introduction

The popularity of grazing in north western Europe is declining (Van den Pol-Van Dasselaar *et. al.*, 2015). Restricted grazing is practised more and more since the average herd size is increasing and many farms have a (very) small grazing platform. To optimise grassland management, farmers need predictions of grass intake of their grazing cows. This study aims to assess grass intake under a range of supplementary silage feeding and stocking densities.

Materials and methods

The whole farm model DairyWise (Schils *et al.*, 2007) was used to calculate grass intake for different stocking densities and different amounts of supplementary silage feeding in the Netherlands. Dairy Wise is a simulation model estimating effects of farm strategies (feeding, fertilising, grassland management) on grassland and animal performance and economics.

First, a baseline was constructed using a farm under good grassland practice with 50 dairy cows, a stocking rate of 1 cow ha^{-1} and day and night grazing without supplementing roughage. The replacement rate was 27% which represents an average Dutch herd including primiparous and multiparous dairy cows. The annual milk production was 8,500 kg milk cow^{-1} with 4.28% fat and 3.47% protein. Annual concentrate supplementation was 2,000 kg cow^{-1}. Second, simulations were carried out for higher stocking densities. This was done by decreasing the grazing platform area by increasing the stocking rates in steps of 0.5 milking cow ha^{-1} and simultaneously increasing the amount of supplementary roughage. The amount of supplementary feed available from own grassland or arable land at farm level depended on the mowing percentage. The mowing percentage on the grazing platform was set between 160 and 230%, which means that the whole grazing platform was mown for silage about twice a year. In the simulations the grassland area was decreased and the amount of supplementing roughage feed was increased until grazing from spring until October was no longer possible. The supplementary feed was a mixture (TMR) of silage maize and silage grass, with a maximum of 5 kg dry matter (DM) silage maize cow^{-1}. Grass silage was harvested on a field paddock or, in case of a shortage, it was bought. For rations with less than 3 kg DM supplementary feed cow^{-1} only grass silage was fed. Thirdly, based on the model output a regression line was estimated with the stocking rate as explanatory factor. The number of grazing hours was estimated based on expert judgement.

Results and discussion

Table 1 shows the results of the model calculations. At low stocking rates (less than 2.5 cows ha^{-1}) cows graze both day and night.

In general the grass intake is 1 kg DM hr^{-1}. Up to 4 kg DM supplementary feed can be fed around milking time. For low grass intake (<4 kg DM) some extra time for grazing was estimated. Figure 1 shows the results of the relation between stocking rate, grass intake and the supplementary feeding. A regression line was fitted using the following equation:

$$\textit{grass intake} = -0.021\times SD^3 + 0.567\times SD^2 - 5.647\times SD + 23.16$$

(where SD = stocking density (cow ha^{-1})).

This function is presented in Figure 1 (fitted grass intake), together with the original data (squares) and the corresponding supplementary feed intake (triangles).

Grazing possibilities at a certain stocking rate depend on the mowing percentage. The less grass is used for silage, the higher the stocking rate can be. But for good grassland practice there should be enough aftermath. Therefore a mowing percentage around 200% was chosen. After every two grazing cuts a cut for silage was planned. Milk production was assumed to be constant. The feed intake sub model is a model based on filling capacity (Delagarde *et al.*, 2011) therefore with maize as supplementary feed instead of grass silage a different grass intake was predicted. In the DairyWise calculations the most common feeding

Table 1. Grass intake per day, grass intake per year (calculated using DairyWise, Schils *et al.*, 2007) and estimated number of grazing hours (expert judgement) for different stocking rates

Stocking rate (cows ha^{-1})	Grass intake (kg DM cow^{-1} day^{-1})	Grass intake (kg DM cow^{-1} yr^{-1})	Estimated number of grazing hours (h)
1	16	2,960	22.0
2.0	14.1	2,584	18.0
3.0	11.0	1,990	12.0
4.0	8.3	1,537	8.5
5.0	6.4	1,201	7.0
6.0	5.4	959	5.5
7.0	3.9	788	4.5
8.0	3.0	664	4.0
9.0	2.6	564	3.5
10.0	2.2	465	3.0
11.0	1.3	345	2.0
12.0	1.0	179	1.5
13.0	0.5	0	0

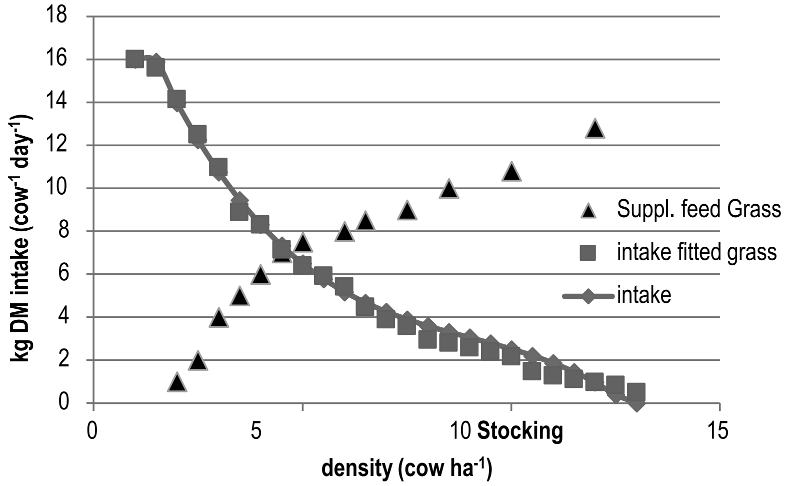

Figure 1. Grass intake fitted (line) and calculated data (squares) and corresponding supplementary feed intake (triangles).

strategy for Dutch circumstances was chosen. The lowest stocking rates in the Netherlands are found on peat soils (CBS, Statline) where maize cultivation is not possible. Therefore the used supplementary feed at low stocking rates in the model calculations was grass silage. This ration better reflects the Dutch situation at low stocking rates. If over 3 kg DM supplementary feed was needed, also maize silage was used in the model calculations.

Conclusions

Predictions of grass intake under a range of supplementary silage feeding and stocking densities on the grazing platform give a quick overview to farmers and advisors of the grazing possibilities on farms. The presented predictions provide a maximum grass intake and the amount of supplementary roughage feed needed at a given stocking rate under good grassland practice.

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Effects of legume establishment by slot-seeding on dry matter and protein yield

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Abstract

According to the demand of public for healthy food, farmers try to avoid genetically varied soya as feed for dairy cows. Increase of protein in roughage therefore should be obtained with higher percentages of legumes in grassland swards. But how can sustainable portions of alfalfa (*Medicago sativa* L.), red clover (*Trifolium pretense* L.) and white clover (*Trifolium repens* L.) be established in permanent grassland? In a field experiment in Oberschwaben (humid climate with best growing conditions for grassland, 5 cuts per year) these species were reseeded in 2012 in two seed rates at two sowing times (early: June; late: August). After three experimental years the percentage of legumes in the sward varied widely and was highest for red clover. Dry matter (DM) yields were numerically highest for red clover in early reseeded treatments. In average no significant effects on DM and protein yields could be reached by reseeding the different legumes in comparison to the untreated control.

Keywords: protein yield, permanent grassland, legumes, *Medicago sativa, Trifolium pratense*

Introduction

The increase of farm-grown protein is a political target in Baden-Wuerttemberg in order to minimize the use of soya in feed rations. Feed protein for milking cows can be produced as farm-grown roughage by a more suitable use of grasslands. High protein yields in permanent grassland can be achieved, among other means, with nitrogen fertilization, early cutting dates or an increase of the legume proportion (Engel *et al.*, 2013; Herrmann *et al.*, 2014). With higher percentages of legumes, the additional waste of fossil energy used during the production of N fertiliser, is avoided. But legumes are not easily maintained because nitrogen fertilization in intensive dairy farming limits the growth conditions of legumes (Luescher *et al.*, 2013). Red clover (*Trifolium pratense*) and alfalfa (*Medicago sativa*) for instance are mainly used as legumes for arable field cropping. Usually they do not tolerate the frequent mowing practiced in intensive grassland systems and their ability for reseeding in permanent grasslands has not been fully proven. Therefore, a field trial was established in 2012 with two sowing dates in South West Germany, Baden-Wuerttemberg. The objective was to prove the practicality of reseeding red clover, white clover and alfalfa in permanent grassland. At the beginning of the experiments, the rates of emergence of the legumes were observed (Engel *et al.,* 2013). Here, dry matter (DM) and protein yields, as well as the development of the botanical composition, of the first three experimental years of the on-going experiments are reported.

Materials and methods

The experimental design follows the description in Engel *et al.* (2013). In a typical grassland region of Baden-Wuerttemberg (Oberschwaben: intensive grass production with 5 cuts; 1000 mm rainfall, 670 m a.s.l.) an experiment on permanent grassland was established.

The treatments described in Table 1 (size of parcels 10m^2, 3 replications) were investigated. As legume species, white clover (*Trifolium repens*), red clover and lucerne (alfalfa) were reseeded at two dates (early: 19 June and late: 23 August). Two seed rates were tested; statistical analysis were done by ANOVA using R, yearwise separately calculated for the two seed dates and mean over the years. To ensure optimum seed establishment conditions, the permanent grassland swards were treated before seeding with herbicide

Table 1. Treatments of legume seed rates, species, varieties and sowing dates.

Factors	Treatments	
Plants and seed rate	Control:	control without reseeding
	WC6:	white clover 6 kg ha^{-1} (varieties: 50% Riesling and 50% Merlyn)
	WC15:	white clover 15 kg ha^{-1}
	RC10:	red clover 10 kg ha^{-1} (varieties: 50% Milvus and 50% Merula)
	RC20:	red clover 20 kg ha^{-1}
	LUC10:	lucerne 10 kg ha^{-1} (varieties: 50% Daphne and 50% Sanditi)
	LUC20:	lucerne 20 kg ha^{-1}
Seed date	Early:	19.06.2012
	Late:	23.08.2012

Starane Ranger 3 lha^{-1} (Triclopyr and Fluroxypyr) in order to avoid uncontrolled establishment of dicotyledons. Additionally, the grassland swards were opened by harrowing in order to allow soil contact of the seed. Legumes were seeded mechanically using the 'Vredo' slot seeder system.The biomass of each cut was dried, weighed, ground and analysed for crude protein content in the years 2013-2015. Prior to each cut, the botanical composition of the sward was estimated for each plot (dry matter yield percentage).

Results

From the start of the experiment on, red clover showed higher percentages than all other legumes. The highest proportions were reached in the 3rd growth in 2014. No significant differences between the sowing rates 10 and 20 kg ha^{-1} were observed by early seed date compared with the late seeding, which showed better development for the higher seed rate of red clover. It was not possible to establish alfalfa neither with early or late seed date nor with different seed rates. White clover could be established and the portions increased in the last growths of each year with a slightly better development by higher seed rate. There is also an increase of legumes in control plots because of spontaneous establishment of white clover (Figure 1 and 2) Due to the experimental design (two separate blocks), the sowing dates were not directly comparable. Nevertheless it seems that it is more successful to sow legumes earlier in the year (Figure 2).

DM and crude protein yields (CPY) differed between the experimental years (Table 2), but there were no statistically significant differences between the treatments except for the early seed date in 2014. However, reseeding with red clover resulted in numerically higher dry matter and crude protein yields on average. It seems that the late sowing date resulted in lower DM and crude protein yields.

Conclusions

Reseeding with red clover in intensively used permanent grassland had, at least occasionally, positive effects on plant establishment, DM and CPY. Reseeding with white clover and alfalfa resulted in lower yields and never leaded to percentages higher than 20%. It could be found that alfalfa was not suitable for improvement of intensive used grassland.

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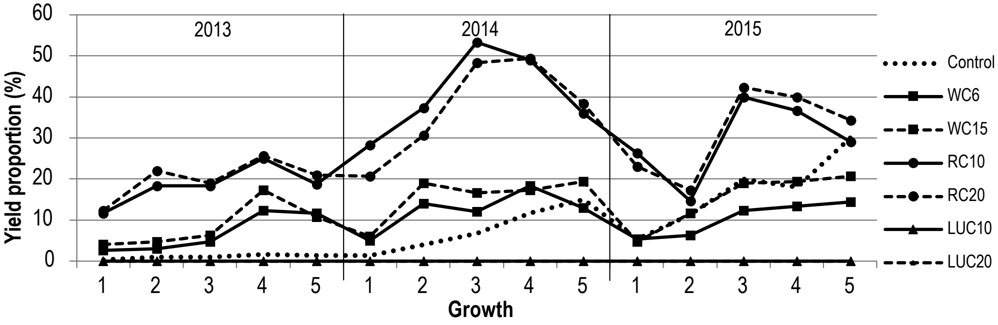

Figure 1. Percentage of sown legumes (%) in each sward from 2013-2015 (early sowing date).

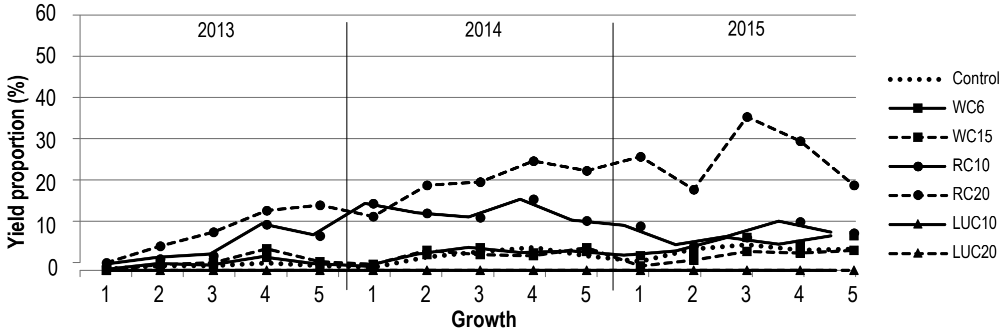

Figure 2. Percentage of sown legumes (%) in each sward from 2013-2015 (late sowing date).

Table 2. Dry matter and crude protein yields (t ha^{-1}) (2013-2015; P=0.05).[1]

		Dry matter (t ha^{-1})				Crude protein yield (t ha^{-1})			
Sowing date	**Treatments**	**2013**	**2014**	**2015**	**2013-15**	**2013**	**2014**	**2015**	**2013-15**
early	Control	9.6a	10.7b	10.3a	10.2a	1.48a	1.73c	1.67ab	1.63a
	WC6	9.2a	10.1b	9.7a	10.0a	1.43a	1.79c	1.52ab	1.58a
	WC15	9.5a	12.7ab	10.6a	10.9a	1.48a	2.02bc	1.65ab	1.72a
	RC10	9.5a	14.1a	11.5a	11.7a	1.54a	2.49a	1.95a	2.00a
	RC20	9.5a	13.9a	11.3a	11.6a	1.51a	2.38ab	1.87ab	1.92a
	LUC10	9.3a	11.9ab	9.9a	10.4a	1.39a	1.89c	1.52ab	1.60a
	LUC20	9.3a	10.4b	10.0a	9.9a	1.38a	1.65c	1.50b	1.51a
late	Control	8.1A	10.4A	9.1A	9.2A	1.17A	1.55B	1.32AB	1.35A
	WC6	9.0A	9.8A	8.2A	9.0A	1.31A	1.44B	1.24B	1.33A
	WC15	9.4A	9.8A	8.8A	9.3A	1.39A	1.52B	1.35AB	1.42A
	RC10	9.6A	11.2A	9.7A	10.2A	1.42A	1.74AB	1.47AB	1.55A
	RC20	8.7A	12.0A	12.1A	10.9A	1.30A	2.02A	1.99A	1.77A
	LUC10	9.0A	10.7A	9.9A	9.9A	1.32A	1.56B	1.45AB	1.45A
	LUC20	9.2A	10.3A	8.6A	9.4A	1.34A	1.59B	1.39AB	1.44A

[1] Different letters show statistical differences; size of letters (a, A) symbolize separate statistical analysis for different sowing dates).

Effect of seeding rate and ryegrass type on sward tiller density and productivity under simulated grazing

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Abstract

The objective of this study was to establish the effect of ryegrass type and seeding rate on sward density and total dry matter (DM) yield under simulated grazing management. Swards with higher tiller densities of ryegrass tend to have greater DM production. Four perennial ryegrass varieties, AberChoice (diploid – D), AberGain (tetraploid – T), Glenveagh (D), Twymax (T) and two hybrid ryegrass varieties, Amalgam (T) and Ligunda (D) were sown in autumn 2013, each at 7.4, 14.8, 22.2, 29.6 and 37 kg ha^{-1}. The swards were simultaneously grazed for the remainder of 2013 (establishment year) with 2014 and 2015 taken as the harvest years. Under simulated grazing management plots were defoliated on nine occasions each year. Plots received nitrogen applications following each harvest totalling 250 kg N ha^{-1} yr^{-1}. There was no seeding rate by variety interaction found on DM yield or tiller density. Seeding rate was found to have no effect on total DM yield but a significant effect on tiller density (P<0.0001). Grass variety had a significant effect on both total DM yield and tiller density (P<0.0001): AberChoice (13.4; 6,015), AberGain (14.2; 3,776), Glenveagh (12.6; 5,511), Twymax (13.3; 4,000), Amalgam (13.3; 3,376) and Ligunda (14.8; 3,336) (t DM ha^{-1}, tillers m^{-2} respectively). These data show that tiller density does not limit variety yield potential within the first 2 full production years.

Keywords: perennial ryegrass, hybrid ryegrass, sowing rate, sward density

Introduction

Perennial ryegrass (*Lolium perenne* L.) tiller density within a sward is shown to impact on DM production (Creighton *et al.*, 2012) but less is understood about hybrid ryegrasses (*Lolium* × *Boucheanum Kunth*) which generally form swards of a lower density. At ploidy level differences in tiller densities of tetraploid and diploid varieties have been noted (Connolly, 2001). Ryegrass sward density is believed to be associated with a plant's ability to persist, and visual ground scores are used within evaluation protocols to measure plant persistence. Persistent swards are highly desirable in Ireland due to the costs associated with a full reseed. Irish farmers aim to achieve swards with a high ryegrass tiller density through variety selection and the use of relatively high seeding rates. As part of the variety evaluation process in Ireland diploid and tetraploid varieties are sown at 30 and 40 kg ha^{-1} respectively, which is representative of seeding rates used on commercial farms (Grogan and Gilliland, 2011). There is poor evidence supporting the use of high seeding rates to achieve dense swards which sustain higher levels of production. The objective of this study was to establish the effect of ryegrass type and seeding rate on sward density and total DM yield under simulated grazing management.

Materials and methods

The study was carried out at Moorepark Animal & Grassland Research and Innovation, Fermoy, Co. Cork, Ireland (50° 09/N; 8°16/W). The soil type is a free draining, acid brown-earth of sandy loam-to-loam texture. Two diploid (D) perennial ryegrass varieties, AberChoice and Glenveagh, two tetraploids (T) AberGain and Twymax plus two hybrid ryegrass varieties, Amalgam (T) and Ligunda (D) were selected as their characteristics indicated that they should form swards of contrasting densities. These varieties were sown in 5×1.5 m plots in autumn 2013, each at 7.4, 14.8, 22.2, 29.6 and 37 kg ha^{-1}. These seeding rates were selected in order to establish a range of contrasting sward densities.

The remainder of 2013 was taken as an establishment phase with measurements commencing in the spring of 2014. These plots were managed under a simulated grazing protocol. In each of the measurement years nine simulated grazings were harvested from the plots. Plots were harvested on a 21-30 day rotation depending on grass growth. Nitrogen fertiliser applications were applied within two days of harvesting. In the spring, mid-season and autumn two turves were removed from each plot and processed to identify the botanical composition of each plot. This generated the ryegrass tiller density of each plot. Visual ground scores were carried out by a team of assessors in December of each year, using the scoring scheme used by variety testing authorities.

This experiment was a randomised complete block design with four replicates per treatment. Six varieties across five different seeding rates were used with plot acting as the unit of measurement. The data from this study was analysed using a mixed model with the fixed effects considered being sowing rate, variety, sowing rate × variety, year × sowing rate, year × variety and block. The dependant variables used in this model were DM yield and tiller density. A 95% confidence interval was set to detect significant differences.

Results and discussion

From the initial two years of this study it was found that ryegrass variety had a significant effect on DM yield and tiller density (P<0.0001): AberChoice (13.4; 6,015), AberGain (14.2; 3,776), Glenveagh (12.6; 5,511), Twymax (13.3; 4,000), Amalgam (13.3; 3,376) and Ligunda (14.8 t DM ha^{-1} and 3,336 tillers m^{-2}, respectively). Seeding rate was shown to significantly influence tillers/m^2 (P<0.0001) but had no effect on DM production (P=0.139). Growing season variation had significant effects on both DM yield and tiller density with yield reducing in 2015 by 1.46 t DM ha^{-1} whilst tiller density increased by 143 tillers m^{-2}. The interactions of seeding rate with year and variety were found not to have significant effects on DM yield or tiller density. The interaction of year and variety did affect DM yield and tiller density.

The results also showed that ryegrass type influenced both DM yield and tiller density. Therefore, variety selection offers a means of establishing swards of higher density as well as higher levels of production and this stabilises regardless of the initial seeding rate. However within the first two full production years, ryegrass sward density does not significantly impact on DM yield. Based on these two years of data the use of high seeding rates as a means of increasing sward productivity is not justified, which is similar to findings in previous ryegrass seeding rate studies (Culleton *et al.*, 1986). Figure 1 demonstrates the effects of seeding rate on sward density and DM yield of swards in their first two full production years.

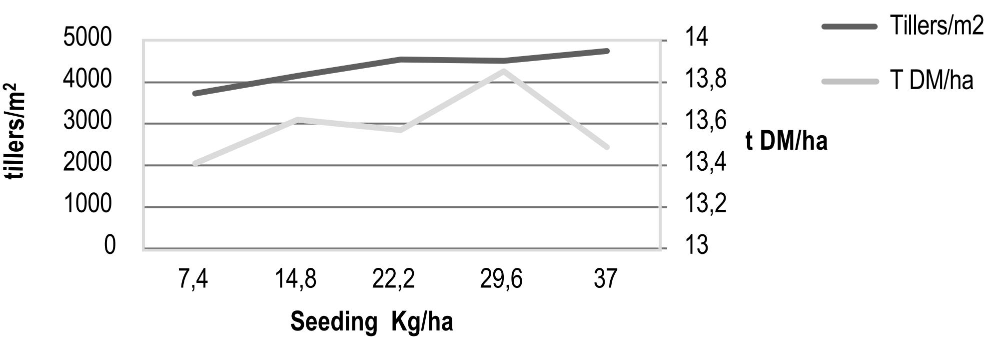

Figure 1. Dry matter yield and tiller density compared to seeding rate.

Conclusions

Swards of lower densities are capable of supporting high levels of production. The indication from visual observation is that this may be due to reduced levels of competition for light and nutrients. It was noticeable that many of the more 'open' swards particularly those of the hybrid varieties contained larger plants that control larger ground and canopy space than plants in more dense swards. To confirm this observation would require an examination of the morphology and levels of light interception of the different varieties across all seeding rates. Given the reported lack of association between density and yield potential, identifying swards in need of renewal should be primarily based on whether DM production is adequate, with sward density only becoming a consideration in terms of animal or machinery carrying capacity. It remains to be determined whether density differences become a more important factor over time as higher density may impact on wear tolerance, the long term persistency of swards and so the economics of reseeding frequency.

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Cattle intake of grass fibres preserved with different additives

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Abstract

the Netherlands is a country with a large area of grasslands. More grass is grown than can be consumed by the livestock. the Netherlands is a substantial protein importer for the feeding of livestock (cows, pigs, poultry). By refining the surplus of the grass, the protein fraction of the grass can be used as a replacement for protein. The grass fibres can be fed as roughage to the calves and cows. This study aimed to test (1) different methods of ensiling grass fibres and (2) the effect of replacing part of the grass silage by ensiled grass fibres. An experiment has been performed at the dairy cattle research center 'Dairy Campus' at Leeuwarden, the Netherlands. The grass refining was done by Grassa! at Gytsjerk, the Netherlands. Based on chemical and feed evaluation analysis combined with the results of the adaptation test with calves, ensiling with molasses was chosen for the performance experiment. The performance experiment shows that grass fibre silage could replace grass silage in the rations of dairy cattle up to 4 kg dry matter cow^{-1} d^{-1} without affecting feed intake and milk production

Keywords: grass, grass fibre, refining, dairy cow feed, Dairy Campus, Grassa!

Introduction

Grass contains a lot of different valuable components, such as proteins, sugars, fibres and minerals. Grass refinery can separate the different components. The protein that is obtained by refining can be fed as replacement of the purchased protein or it can be sold and fed to poultry and pigs, thereby replacing imported protein. The grass fibres can be used as roughage for the cattle and replace the grass silage. This way, the cattle farmer adds value to the grass. The current knowledge on ensiling grass fibres and feeding grass fibres is limited. Therefore two items are of importance: (1) it is necessary to carry out research into the best way of ensiling grass fibres and (2) the effect of replacing grass silage by grass fibres on the feed intake and the milk production.

Materials and methods

The research consists of two parts: a preservation test combined with an adaptation test with calves and a performance trial with dairy cows. Until now silage quality and feed evaluation of grass fibres was unknown. Therefore the fresh grass fibres were ensiled with different additives and an adaptation test has been performed. The performance trial for dairy cows has been performed to investigate the effect of the milk production with ordinary silage and with the replacement of refined and low-protein grass fibre. Firstly, the effects of different additives to ensiled grass fibres were tested. For each treatment 15 kg of fresh grass fibres (fgf) were mixed with the following additives: molasses (in 2 quantities: 40 and 80 g kg^{-1} fgf); molasses + Ecosyl 50 (80 g molasses and 2 ml kg^{-1} fgf Ecolsyl solution 2 g l^{-1}); Ecosyl 50 (2 ml kg^{-1} fgf Ecosyl solution 2 g l^{-1}); propionic acid (3 g kg^{-1} fgf); salt (NaCl, 2 g kg^{-1} fgf); Na-glutamate (5 g kg^{-1} fgf); NaOH (3 g kg^{-1} fgf); and urea liquid (5 g kg^{-1} fgf). All ensiled grass fibres have been fed to Holstein-calves and were analysed on chemical and feed evaluation parameters. Results of the analysis are not mentioned in this paper. All silages were offered simultaneously in a preference test to the calves. After an hour the remainder of the silages were weighed. The intake was calculated as percentage of the total fibres offered. The test was not statistically analyzed.

Secondly, the effect of replacing part of the grass silage by ensiled grass fibres on feed intake and milk production of dairy cows was tested using grass fibres ensiled (gfs) with molasses (80 g kg^{-1} grass fibre). The grass fibre silage had been ensiled in round bales. Two groups of each ten cows were used. The test lasted five weeks. There were two treatments with 10 cows each where part of the grass silage was replace by grass fibre silage. The first week both groups had 2 kg dry matter (DM) grass silage replaced by grass fibre silage cow^{-1} d^{-1} in the ration. For group A (low gfs) the amount of grass fibre silage decreased to 0 kg in week 3 and then increased to 2 kg DM in week 5. For group B (high gfs) the amount of grass fibre silage increased to 4 kg in week 3 and then decreased to 2 kg DM in week 5. The cows received a total mixed ration of grass fibre silage, grass silage, maize silage, soybean meal and molasses syrup at the feeding pitch and concentrates (5.5 kg cow^{-1} d^{-1}) in the milking shed. Feed intake, milk production and ruminating behaviour was measured and faeces quality was evaluated.

Ruminating activity was continuously measured with all the cows using sensors on the collar (Lely Qwes-HR). In each trial week, from at least half the number of cows a faeces sample was taken. In the faeces the dry matter content and the fibre fraction was determined. In order to determine this fibre fraction, the faeces sample was rinsed out over a household sieve (mesh size 2 mm). The coarser faeces particles remaining on the sieve were dried, weighed and expressed as % of the total amount of faeces, prior to screening. The feed intake, milk production results and other observations from this trial have been statistically tested with a REML model with random effects group of animals (animal effects) and trial week (period effects) and as fixed effects linear trend test week (cows recovered after all, lactation stage) and the estimated impact of grass fibre gift (within trial week).

Results and discussion

Figure 1 shows the feed intake of ensiled grass fibres ensiled with different additives. Silages ensiled with molasses (low quantity) and propionic acid scored best on feed intake. The disadvantage of using propionic acid is that the acid leads to corrosion on ensiling equipment.

The silages with urea and NaOH were not eaten at all. The intake of the other silages (including the control silage without additive) varied between 40% and 60%. Based on chemical and feed evaluation analysis combined with the results of the adaptation test with calves, ensiling with molasses was chosen for the performance experiment.

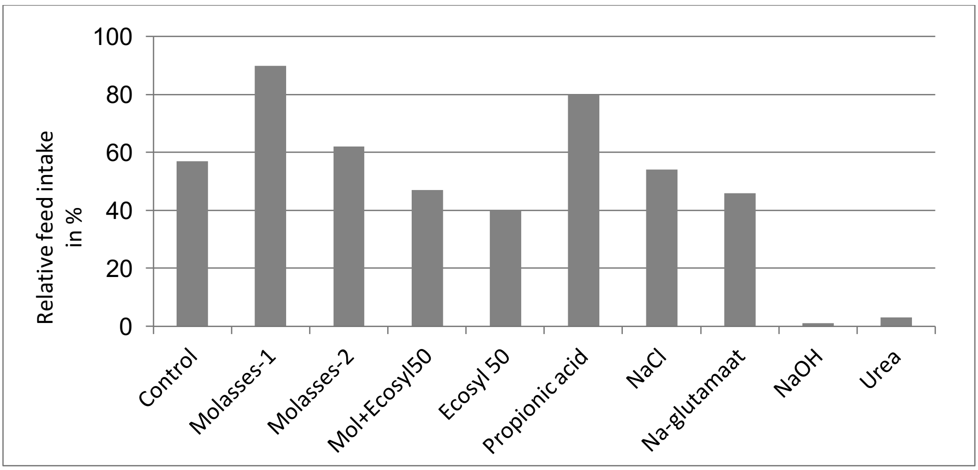

Figure 1. Relative feed intake (in % related to the feed gift) in the intake test with calves.

Replacing grass silage by grass fibre silage did not significantly effect the feed intake of dairy cows (20.8 and 20.3 kg DM cow^{-1} d^{-1}, for the grass silage group and the grass fibre silage group, respectively) (P=0.751). Furthermore, there was no clear effect on milk production (Figure 2).

Ruminating activity was measured and the average ruminant time was 21 min per hour, which corresponds to 35% of the total time. There was no statistical difference between the two groups over the trial period. The ruminating activity of the cows did not change when grass fibre silage was provided. Certainly, the dry matter percentage of the faeces was slightly lower (P<0.041), the more grass fibre was feeded. However, in this test it did not result in a different digesting of the ration, as it was expressed in the fibre fraction in the faeces.

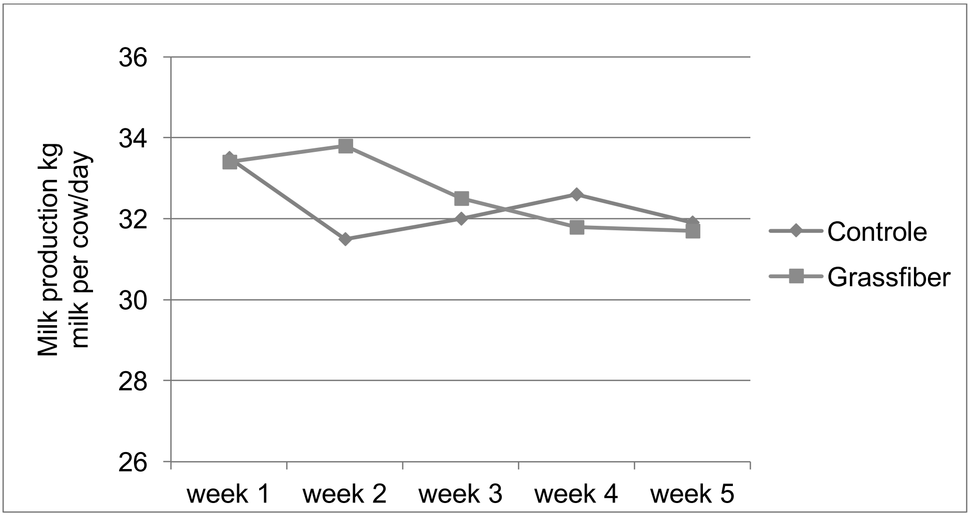

Figure 2. Average day produce in kg milk per group, per test week (kg milk per cow/day)

Conclusions

Intake of grass fibres was highest for grass fibres that were ensiled with molasses. Grass fibre silage could replace grass silage in the rations of dairy cattle up to 4 kg DM cow^{-1} d^{-1} without affecting feed intake and milk production.

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Utilising common vetch (*Vicia sativa*) as a source of forage protein for grazing ewes

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Abstract

Integrating high protein forages into livestock systems is fundamental to the sustainable development of farming. Typically, the winter period is the most difficult time to produce a home-grown forage protein source as a replacement for purchased feed. During lactation, ewes have a high demand for high quality forage to maintain milk supply. To address this, both a controlled replicated experiment (Aberystwyth University) and participatory research (commercial development farm) were conducted to investigate the effects of common vetch (*Vicia sativa*) when grown over the winter period and offered to lactating ewes. The findings from the controlled research showed that feeding a vetch/Italian ryegrass mix to lactating ewes resulted in a higher combined ewe and lamb weights but only on Day 35. Participatory research indicated that common vetch had a useful role in providing forage protein to post lambing ewes in commercial farming practice, was able to regrow for several grazing periods and offered an opportunity to reduce N fertiliser. Overall, both studies showed there were no detrimental effects of feeding vetches to sheep and that vetch has the potential to be used as a source of high-protein fresh forage in spring to support lactating ewes and their lambs.

Keywords: *Vicia sativa,* legumes, forages, protein, sheep, lactation

Introduction

Utilising high protein forages within ruminant livestock systems is fundamental to the sustainable intensification of farming systems globally to reduce reliance on imported protein. Common vetch (*Vicia sativa*) is a leguminous forage which is often used as a cover crop to suppress weeds and build soil fertility and is mostly sown as a bi-crop in combination with either barley or oats (Dhima *et al.*, 2007). Typically, farmers find the winter period to be the most difficult time to produce a home-grown forage protein source to reduce the requirement for purchased concentrate feed in spring. Coupled with this, lactating ewes during the spring lambing period have a high demand for high quality forage to maintain milk supply, with their intakes of dry matter increasing up to 3.5% of total body weight in early lactation. These two factors often result in a need for concentrate feed to be purchased to meet the nutritional needs of the ewe during early lactation. To address this, (1) a controlled replicated experiment (Aberystwyth University) and (2) participatory research (on a commercial development farm) were conducted to investigate the effects of vetch, grown over the winter period, and offered to lactating ewes as a protein feed at pasture.

Materials and methods

Controlled replicated experiment. The experimental design comprised 2 replicate plots of two treatments, with 8 ewes, each rearing twin lambs, grazing each plot. The grazing experiment ran from mid-April to mid-June 2015 and measured the performance of ewes and their lambs for 7 weeks post-lambing. Experimental plots (approx. 0.8 ha) were sown with either (1) a common vetch (*V. sativa*, cv. Slovena)/Italian ryegrass (*Lolium multiflorum*, cv. Dorike) mix (Vetch/IRG) at 24.7 and 24.7 kg ha^{-1} respectively) or (2) with Italian ryegrass (cv. Dorike) (IRG) at 34.6 kg ha^{-1} on 19 September 2014. Mule ewes selected from a flock (n=114) were mated in 3 groups starting 17 November using 3 Texel rams. All ewes lambed indoors (commencing 12 April) and were fed according to scanning results and following standard farm practice. All twin-rearing ewes were moved outdoors at 24 h post-lambing and kept as one group on a grass pasture and offered standard ewe concentrate (at 400 g head^{-1} d^{-1}) until allocated to plots on the basis of lambing date, ram group at mating and lamb birth weight. During the grazing experiment, ewes did not receive supplementary concentrates. Forages were maintained in a vegetative state by grazing rotationally across each plot using electric fencing and back-fencing moved weekly to allow regrowth. Above ground forage biomass was determined within three 0.5×1 m quadrats, cut within each plot at the start and end of each week. Dry matter (DM) content was determined on a sample from each quadrat, bulked and a composite subsample was taken for quantification of proportions of sown IRG, vetch, weed grass and broadleaf weed on a DM basis. Ewe and lamb live weights were recorded weekly as animals were moved. Forage data were analysed by ANOVA and animal live-weight gain within each experimental period was calculated as the difference between the final and start live weight data. Combined ewe and lamb liveweight data were compared by ANOVA using repeated measures.

Participatory research. Linked to the controlled experiment at IBERS, on a sheep commercial development farm in SW England, the participatory research explored the role of vetches in a catch crop of hybrid ryegrass with the aim of increasing the grazed forage protein offered to lactating ewes with lambs in early spring. At a field scale, the catch crop ryegrass with and without vetch was monitored by the farmer during the growing seasons 2012/2013 and 2013/2014. Common vetch (cv. Slovena) broadcast at 24.7 kg ha^{-1} and hybrid ryegrass (*Lolium hybridum*) (cv. AberEcho) and Festulolium braunii (*Lolium boucheanum*) (cv. Aberniche) broadcast at 12.35 kg ha^{-1} each respectively, following a cereal within a rotation.

Results and discussion

Controlled replicated experiment. Forage data showed ewes selectively grazed vetch in the Vetch/IRG treatment (Table 1) and that vetch re-grew over 21 days when rotationally grazed (data not shown). Ewe liveweight changes were within the range typically reported for ewes in the first few weeks post-lambing (Cowan *et al.*, 1980). There was no effect of forage treatment on ewe or lamb or combined ewe and lamb liveweight changes (Table 1). A limitation of this work was the number of replicated plots, 3 replicates were established for grazing but one replicate of each treatment was found to be unsuitable for inclusion. However, on Day 35, the combined liveweight of ewes and their lambs on the Vetch/IRG treatment was higher than that of those on the IRG only sward (P<0.05) (Figure 1). This finding suggests that

Table 1. Average total forage, vetch and Italian rye grass (IRG) yield (kg dry matter ha^{-1}) available to ewes with twin lambs and liveweight gain of ewes and/or lambs grazing either Vetch/IRG or IRG only.

	Vetch/IRG	IRG	sed	Prob
Pre-grazing yield (kg dry matter ha^{-1})				
Total	1,913	1,917	37.0	0.914
Vetch	235	–	–	–
IRG	1,241	1,453	51.4	0.054
Post-grazing yield (kg dry matter ha^{-1})				
Total	1,201	1,390	154.8	0.346
Vetch	31	–	–	–
IRG	885	1,011	141.2	0.466
Liveweight gain				
Ewe (g/d)	-74	-98	53.3	0.694
Lambs (g/2 lambs/d)	610	563	33.5	0.296
Combined (g/ewe+2 lambs/d)	579	477	61.1	0.235

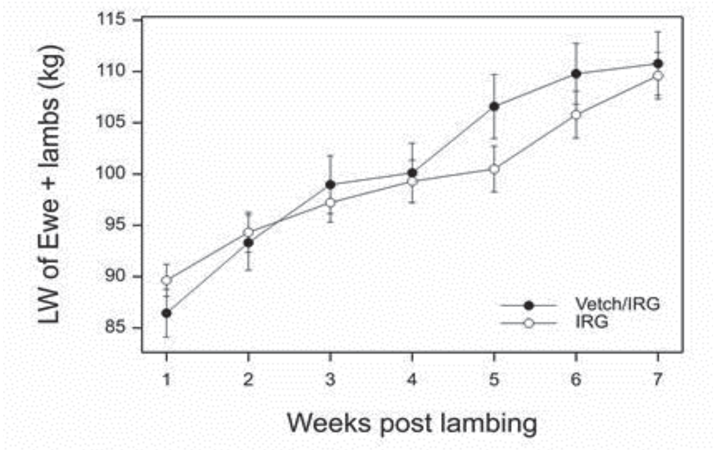

Figure 1. Combined liveweight of ewes and twin lambs grazing either Vetch/Italian rye grass (IRG) or IRG only.

improvements in performance of lactating ewes were made from offering fresh vetch to ewes post-lambing but that these benefits required time to become apparent, possibly due to the period needed for dietary adaptation changes in the rumen.

Participatory research. Farmer estimates of the yield indicated that, although the grass with and without vetch had a mean of 9.9 t DM ha^{-1}, the yield achieved by the grass alone had received an additional 86.25 kg N ha^{-1} of fertiliser. The crude protein (%) levels of the grass alone averaged 25.8 compared with 27.3 for vetch and grass. The ewes and lambs rotationally grazed the area from March to July allowing regrowth of grass and vetches. The farmer reported that once established vetch grew rapidly and vigorously and regrew after grazing.

Conclusions

Feeding a Vetch /Italian ryegrass mix to lactating ewes post-lambing resulted in a higher combined ewe and lamb weights but only on Day 35. Participatory research indicated vetch had a useful role in providing forage protein to lactating ewes and reducing N fertiliser use in commercial farming practice. Overall, there were no detrimental effects of feeding vetches to sheep and vetch has the potential to be used as a source of high-protein fresh forage in spring to support lactating ewes and their lambs.

Acknowledgements

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Direct sowing of red clover by three technologies

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Abstract

At Vatín site in the Czech Republic, an accurate small plot trial was established in 2011 on two different sowing dates (June and July) with three technologies, consisting of directly sown seeds of red clover 'Amos' (4n) and 'Suez' (2n). Direct seeding was carried out by the technologies of slot seeding with a seeding drill SE-2-024, and strip seeding with a prototype of PP-2 drill and a prototype of STP 300 drill. Radical disturbance (PP-2) of the original sward was associated with a higher (P<0.05) dry matter yield and was reflected in higher (P<0.05) dominance of direct sown species (38% and 52%) in the first year. The direct sown species persisted better with the SE-2-024 and STP-300 technologies, which were less radical.

Keywords: direct sowing, red clover, grasslands, production, quality, persistence

Introduction

Introduction of direct sowing into grasslands is influenced by a number of factors. The successful use of direct sowing is still low and therefore more research, development and verification of new technologies is required (Kohoutek *et al.,* 2002). Technologies of additional sowing differ according to the rate of the original grass sward disturbance. Treatments with rotavator may give narrow gaps or broader furrows or affect the entire original grass sward. Another sowing possibility is a mere spreading of seeds with the subsequent use of a land leveller (Skladanka *et al.*, 2012). The aim of this research was to evaluate the yields of direct sown stands and projective dominance of red clover after using different technologies on two different sowing dates. The consequential aim is the evaluation of forage quality of direct sowing grasslands.

Materials and methods

At Vatin site in the Czech Republic (average annual temperature 6.9 °C, annual long-term rainfall average 617 mm, altitude 553 m, a multifactor experiment with direct sowing into permanent grassland (PG) was established in 2011. The meadow vegetation is represented by a plant society consisting of *Arrhenatheretum*. Three direct sowing technologies were used: (1) slot seeding with SE-2-024 (rotary coulter, width of the grooves 40 mm), (2) strip seeding with PP-2 (radical disturbance, width of the grooves 150 mm) and (3) strip seeding with a prototype of STP 300 drill (discs with coulters) with spike seeding mechanism. The trials were established in two time arrangements (17 June – T1 and 18 July – T2). Red clover 'Amos' (4n) and 'Suez' (2n) were sown at a rate of 35 kg ha^{-1}. The plot length was 16.11 m, the length of harvested part was 11.11 m, with 3 replications.

The trial plot was not fertilized in the year of direct sowing (2011). In the first and second harvest year (2012 and 2013) the plot was fertilized at rates of 35 kg ha^{-1} P and 100 kg ha^{-1} K. Nitrogen was only applied on directly sown grasses at the rate of 180 kg ha^{-1} N (60-60-60) in the nitrate form, whilst the variants with directly sown legumes were without nitrogen fertilization. The trial plot was harvested by three cuts. This paper evaluates dry matter yield (sum of three cuts) and projective dominance (in the first cut). The forage quality (in the first cut) was measured using near infrared spectrometry (NIRSystems 6500). The observed parameters were: crude protein (CP), crude fibre, net energy of lactation. The measured results were statistically evaluated and differences between averages were tested with the Tukey test.

Results and discussion

Dry matter yield in the first year after direct sowing ranged from 6.89 to 10.05 t ha^{-1}. Radical disturbance (PP-2) of the original sward was associated with a higher (P<0.05) dry matter yield. The differences (P<0.05) were also evident in the dominance of direct sowing legumes (Table 1).

Radical disturbance of grass sward was reflected in higher (P<0.05) dominance of direct sowing species (38 and 52%). In the second year after sowing, the yield of direct sown grasslands normalized and ranged from 9.66 to 10.43 t ha^{-1}. A difference in the dominance of sown species was evident (P<0.05). Sown legumes dominated with technologies SE-2-024 (60%) and STP-300 (39%). By contrast, the dominance of red clover declined in the stands with technology PP-2 (26%). The results show that the radical disturbance sward (PP-2) significantly influenced the yield in the first year after direct sowing (P<0.05). The direct sown species persisted better in the technologies SE-2-024 and STP-300, which were less radical. However, it is always a technology delivering seed into the soil. Decrease in clover dominance from 2012 to 2013 (PP-2) was related to facilitation for grass growth and dominance by the great N input from clover N-fixation in 2012. However, the low N input in 2012 (SE-2-024 and STP-300) facilitated clover relative to grass in 2013 (Table 1).

The date of sowing did not affect yield, and is reflected in the dominance of direct sown species. The first date of sowing has supported dominance of red clover (Table 1). Although the technology has affected the yield of grassland and the dominance of sowing red clover, there was not a considerable difference in the quality of forage (Table 2). The difference among the technologies is particularly evident in the second year after sowing, when the technology SE-2-024 led to a higher (P<0.05) content of CP (150.25 g kg^{-1} DM). This technology has been associated in the second harvest year (2013) with the highest dominance of direct sown species.

Table 1. Influence of the variety, technology and date on the yield of dry matter (DM) (t ha^{-1}) and projective dominance (%) in the years 2012 and 2013.[1]

Factor	2012		2013	
	DM yield (t ha^{-1})	Dominance of red clover (%)	DM yield (t ha^{-1})	Dominance of red clover (%)
Variety				
Amos (4n)	8.67	39	10.24	49
Suez (2n)	8.24	30	9.91	34
Technology				
SE-2-024	8.42^{ab}	38^{a}	10.43	60^{a}
STP-300	6.89^{a}	14^{b}	9.66	39^{ab}
PP-2	10.05^{b}	52^{a}	10.15	26^{b}
Date				
D1	8.80	48^{a}	9.83	40
D2	8.11	21^{b}	10.33	43

[1] Average values in the same columns with different superscripts are statistically significant at a level of P<0.05.

Table 2. Influence of the variety, technology and date on the content of crude protein (CP) and crude fibre (CF), (g kg^{-1}) and content of net energy lactation (NEL), (MJ kg^{-1} dry matter) in the first cut of the years 2012 and 2013.[1]

Factor	2012			2013		
	CP (g kg^{-1})	CF (g kg^{-1})	NEL (MJ kg^{-1})	CP (g kg^{-1})	CF (g kg^{-1})	NEL (MJ kg^{-1})
Variety						
Amos (4n)	134.65	233.09	5.88	137.47	273.16	5.45
Suez (2n)	129.63	236.9	5.85	132.10	275.32	5.45
Technologies						
SE-2-024	127.40	244.40	5.74^{a}	150.25^{a}	257.27	5.58
STP-300	135.32	233.45	5.92^{b}	124.30^{b}	282.07	5.39
PP-2	133.71	227.20	5.93^{b}	129.80^{ab}	283.37	5.39
Date						
D1	140.84	226.31	5.90	135.61	269.77	5.42
D2	123.44	243.73	5.83	133.96	278.71	5.48

[1] Average values in the same columns with different superscripts are statistically significant at a level of $P<0.05$.

Conclusions

Yields of stands, as well as the dominance of sown species, was affected by direct sowing technology. Radical disturbance of the sward (PP-2) led to higher yield and higher projective dominance of direct sown legumes in the first harvest year. The difference in yields levelled off in the second harvest year. By contrast, the dominance of sown legumes persisted especially with technologies SE-2-024 and STP 300. Higher dominance of red clover in the second harvest year with technology SE-2-024 was also reflected in higher CP content in the forage.

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Theme 4.
Synergies between ecosystem services, biodiversity and agricultural production in grasslands

Ecosystem service indicators for grasslands in relation to ecoclimatic regions and land use systems

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Abstract

Since the Millennium Ecosystem Assessment, the concept of ecosystem service (ES) has been increasingly used by scientists and policy makers. For example, the European Union launched an extensive mapping program of ES mapping in all Member countries. The objective of this paper is (1) to examine the relevance and feasibility of the assessment of ES delivered by grasslands using indicators, and (2) to question whether it is necessary or not to adapt them to ecoclimatic regions and land use systems. Firstly, we show that the concept of ecosystem service has not achieved consensus, which complicates its implementation. Based on case studies in different European regions (Continental, Atlantic, Mediterranean, Nordic), and on results from European research projects which have referred to the development of ES indicators, we show the diversity of possible approaches. This diversity concerns various aspects: the objectives of the indicators, the beneficiaries of the ES, spatial and temporal scales, and organization levels. In conclusion, we think that it is necessary to develop ES indicators adapted to ecoclimatic and land-use conditions. The issue of ES is a global issue, and thus it appeared desirable to find common indicators in order to compare the systems, but to adapt the calculation to specific conditions.

Keywords: ecosystem services, grassland, indicator, case study

Introduction

It is now clearly established that grasslands and grassland-based livestock systems play an essential role in the delivery of many ecosystem services (ES). For instance, the key role of semi-natural grasslands in delivering ES was demonstrated by the UK National Ecosystem Assessment (UK NEA, 2011). Grasslands in Europe cover some 35% of the agriculturally utilized land area. However, over the last four decades the overall grassland area in Europe has been declining, with tangible implications on biodiversity and the delivery of ES like C sequestration, biodiversity conservation or erosion control (Huyghe *et al.*, 2014). Human benefits from all services associated with semi-natural grasslands have either declined or show a mixed trend because the number, biodiversity value and size of semi-natural grasslands have dramatically declined in Europe (Harrison *et al.,* 2010). For example, the European Grassland Butterfly Indicator, which assesses the size of butterfly populations, calculated by the European Environment Agency decreased by 50% between 1990 and 2011. Evaluating the value of grassland for society, in particular the costs of grassland losses or degradation, requires indicators. This issue is complex for several reasons: (1) because the concept of ecosystem service is fairly recent, and its definition evolves regularly and is not shared by all, (2) because different objectives might require the assessment of the delivery of ES at different scales (grassland, farm and/or territory level), and (3) the development of indicators both scientifically valid and relevant for their users is a complex process. Finally, it will be key not only to select relevant indicators of ES but also to establish whether they can be the same for the different situations (ecoclimatic regions, land-use systems) encountered across Europe.

The aim of this paper is to analyse the concept of ES, to identify ES delivered by grasslands and indicators for the assessment of ES, to evaluate their applicability while employing examples (case studies), and discuss future needs of research and development of indicators

The concept of ecosystem services

The Study of Critical Environmental Problems (SCEP, 1970) can be seen as the reports addressing 'environmental services' like pollination, climate regulation, flood control, and soil erosion prevention. Westman (1977) in his work on 'nature's services' provided a first detailed analysis of several key 'service(s) of an ecosystem'. While questioning the possibility of valuation of these services, he pointed out the necessity of quantitative estimates to highlight the 'dependence of human civilization on the services provided by ecosystems'. In their book 'Extinction', Ehrlich *et al.* (1981) clearly highlighted the consequences for mankind of species extinction: failure of crops, severe pests outbreaks, loss of soil fertility, freshwater supply, danger of floods, ... They stated that 'Natural ecosystems maintain a vast genetic library that has already provided people with countless benefits and has the potential for providing many, many more'.

After Ehrlich *et al.* (1981), the two seminal works of Daily (1997) and Costanza *et al.* (1997) brought to the spotlight the concept of ES. Notably, these two key authors defined ES differently. While Daily (1997) considered ES as 'conditions and processes associated with natural ecosystems that confer some benefits to humanity' Costanza *et al.* (1997) considered them as 'benefits human populations derive, directly or indirectly, from [...] habitat, biological or system properties or processes of ecosystems'. In other words, the first consider ES as 'conditions or processes' of an ecosystem while the second consider them as benefits derived from these conditions or processes. This ambiguity in ES definition still remains today (Fisher *et al.*, 2009; Kandziora *et al.*, 2013; Villamagna *et al.*, 2013; Wallas, 2007). However Daily's conceptualisation seems to be the one that is primarily featured in recent international projects like the development of the 'Common International Classification of Ecosystem Services – CICES' (EEA, 2013), the SEEA-EEA (UN, 2013), the European programme 'Mapping and Assessment of Ecosystems and their Services – MAES' (Maes *et al.*, 2013). This conceptualisation has been strongly supported by the often-cited work of Fisher *et al.* (2009) in which "ecosystem services' are the aspects [structure or processes] of ecosystems utilized (actively or passively) to produce human wellbeing'. The recent development of CICES helps to clarify the concept of benefits: they are derived from ecosystem services and most often require integration of other forms of capital (human, social, manufactured and financial, see also Palomo *et al.*, 2016). In contrast to ES, benefits are no longer functionally connected to the ecosystem (see EEA, 2013 and UN, 2013).

The Millennium Ecosystem Assessment report (MEA, 2005) strongly influenced the dissemination of the ES concept. According to MEA, like Costanza *et al.* (1997), ES are benefits for the people obtained from ecosystems. These benefits are classified into provisioning, regulating, supporting and cultural services and they apply to natural or semi natural ecosystems as well as to systems that are stronger modified and controlled by human. ES cover all aspects of human well-being. Costanza *et al.* (1997) and many others (e.g. Barnaud and Antona, 2014; Costanza, 2008; Costanza *et al.*, 2014; De Groot *et al.*, 2012; Fisher *et al.*, 2008) highlight that most of ES are public goods (non-rival and non-excludable) or common pool resources (rival but non-excludable). Huang *et al.* (2015) explain that some ES correspond to non-marketed functions of the multifunctional agriculture framework. Accordingly many authors (e.g. Costanza *et al.*, 1997, 2014; De Groot *et al.*, 2012; TEEB, 2010) argue that ES should be valued in money terms so that their contribution to human economy can be considered and compared to marketed goods and services. Estimating a 'total economic value' integrating all components of utility derived from ecosystem services is a way to perform such monetary evaluation (e.g. Costanza *et al.,* 1998; De Groot *et al.*, 2012). Identifying ES and their role in supporting human life, understanding the way how they are generated and knowing trade-offs among services within socio-ecological systems is the prerequisite for the maintenance of the services. This will then form the basis to guide economic and land-use activities of ES utilization (Bateman *et al.,* 2013; Sanderson *et al.*, 2007), to support political decision making (Diehl *et al.,* 2016; Fisher *et al.,* 2008; Förster *et al.,* 2015; Loft *et al.,* 2015), and to define research needs and design research approaches (Bennett *et al.,* 2015; Seppelt *et al.,* 2013). However, while Daily (1997) and Costanza *et al.* (1997) distinguished ecosystem goods from ES, the MEA (2005) and most of the works after, consider both under the umbrella of 'ecosystem services'. This simplification led to a consideration that all goods provided by ecosystems are ES. This conceptualisation is very problematic when dealing with very anthropized ecosystems, where goods correspond to a co-production of anthropogenic capital (built, human, social) and ES (Albert *et al.*, 2015; Bengtsson, 2015; Bommarco *et al.*, 2013; Duru *et al.*, 2015a; Palomo *et al.*, 2016; SEEA-EEA, 2013). Following on from Daily (1997), Nelson and Daily (2010) specified that ES include processes that support the production of consumable goods (e.g. food and timber), processes that support and regulate life (e.g. crop pollination and carbon sequestration), conditions that enhance life (e.g. recreational, aesthetic landscape), and conditions that preserve valuable options (e.g. potential benefits from biota). In line with this definition, focusing on agriculture, Bommarco *et al.* (2013) claim that 'it is important to make the distinction between services as extracted goods [...], or as underpinning processes' (see also Fisher *et al.*, 2009).

Zhang *et al.* (2007) and Swinton *et al.* (2007), analysing interactions between ES and agriculture, highlighted that agriculture both provides and receives ES. Le Roux *et al.* (2008) in their analysis of interactions between biodiversity and agriculture called 'inputs services' the ES provided to agriculture. In this conceptual perspective, Bommarco *et al.* (2013) and Duru *et al.* (2015a), clarified the status of ES in agricultural production. As with others (e.g. Garbach *et al.*, 2014), they highlight that regulation services that determine soil fertility (soil structure and nutrient cycling) and pests control are the key ES provided by ecosystems to agriculture. Developing ecosystems that provide a high level of these ES can enable farmers to significantly decrease use of exogenous inputs. Duru *et al.* (2015a) established clearly the link between the well-known agronomic theory of defining, limiting and reducing-growth factors of Ittersum and Rabbinge (1997) and the theory of ES provided to agriculture. As with Bommarco *et al.* (2013), they show that the agricultural production (or goods) is more or less determined by anthropogenic exogenous inputs and ES. In other words, the share of the agricultural production depending on ES (vs exogenous inputs) is more or less important according to the nature of farming systems: input-based or ecosystem service-based farming systems (Duru *et al.*, 2015a). It is however important to keep in mind that even input-based farming systems depend on ES (Bommarco *et al.*, 2013; Duru *et al.*, 2015a), and they could be considered as ES that are transferred from other locations.

Grasslands correspond to various agroecosystems that can be more or less intensive, i.e. in which forage production depends more or less to exogenous inputs or ES (soil fertility, biological regulations including pollination). As has been conducted for agricultural ecosystems in general (e.g. Garbach *et al.*, 2014), ES provided by grasslands to farmers and society have been recently investigated (e.g. Duru *et al.,* 2015b; Rodríguez-Ortega *et al.,* 2014). The abundance and spatial distribution of grasslands in farmlands and landscapes as well as their temporal distribution in crop sequences strongly determine soil fertility and biological regulations as well as water, climate mass, water and air regulation and cultural services (Lemaire *et al.*, 2015; Moraine *et al.*, 2016; Soussana and Lemaire, 2014).

Evaluation of ecosystem services

Many ecosystem service assessment approaches have been developed to quantitatively assess ES. They allow analyses of the spatial configuration of ES and in turn can support land use planning and ecosystem management (Malinga *et al.*, 2015; Martínez-Harms and Balvanera, 2012; Seppelt *et al.*, 2011). The most common methods use 'process-based' models fed with secondary data linked to land cover, land use and soils (Martínez-Harms and Balvanera, 2012). Most are applied at 'intermediary level' (municipality and province) with a fine spatial resolution of one ha or less (Malinga *et al.*, 2015). Despite important progress most approaches are still under development (Kandzioraetal., 2013; Malinga *et al.*, 2015; Seppelt *et al.*,2011). In these approaches the spatiotemporal dynamics of ES is still poorly assessed (Birkhofer *et al.*, 2015; Oliver *et al.*, 2015; Renard *et al.,* 2015; Wood *et al.*, 2015). Approaches have been developed to assess how multiple ES are bundled together within a studied extent (e.g. Raudsepp-Hearne *et al.*, 2010) and these allow assessments of landscape multifunctionality, i.e. 'the joint supply of multiple ES at the landscape level' (Mastrangelo *et al.*, 2014). At present, most of the available approaches are being developed and tested by scientists only and are 'pattern-based'. Thus, more stakeholder involvement and further studies on the processes linking different ES are required in order to develop 'socially-relevant process-based approaches', which would be both relevant for local stakeholders and flexible in their utilization. Such approaches are based on involvement of local stakeholders all along the process from the identification of socially-relevant ES to the analyse of socially-relevant ES bundles (*ibid.*).

As highlighted by Duru *et al.* (2015a), ES are scale-dependent and most of them are studied at field and landscape levels, whereas the farm level is notably under-studied even though it is the level at which land-use and management decisions are made. The focus on ES delivered by grasslands can therefore give a biased picture, and the whole ecosystem of production systems must also be taken into account. In this regard, an important question is to define the situation of the farmers: can they be considered as intermediaries between ecosystems and the public, or are they as manipulators of ecosystem processes that modify the 'natural' flow of ES from the natural world?

After Westman (1977), the question of ES evaluation was taken into account by economists, and interesting estimations were performed like in Czech Republic in a survey of ecosystem services of the main types of grasslands (Table 1; Hönigova *et al.*, 2012). The ecosystem accounting of grassland ES has been addressed in this pilot study on grasslands in the Czech Republic based on a habitat ecosystem accounting approach and value/benefit transfers (Hőnigová *et al.,* 2012). While grassland ecosystem are usually accounted within a single ecosystem category, habitat accounting enables differentiation and enables more detailed classification of ecosystem services flowing from habitats with different characteristics.

Concerning the economic valuation of grassland ecosystem services, appropriate methods are still being tested. For example, for livestock production, production function should be constructed which would give estimate of ecosystem contribution to marketed value. In many cases, the economic values are based on proxies or benefit transfers. Proper valuation of grassland ecosystem services thus still remains a challenge. However, economic valuation enables the quantification of ecosystem services contributing the economic production and income.

Ecosystem services delivered by grasslands

Due to the diversity of definitions of ES, it is not surprising to find in the scientific literature and in the political world very many ways to evaluate the ES. In this text, we chose not to select a single definition, but to report on the different possible approaches, ES as 'conditions or processes' of ecosystem or benefits derived from these conditions or processes.

Table 1. Summary of calculated monetary values of selected grassland ecosystem services (Hönigova *et al*., 2012).

Code	Category	Max livestock number (Eur ha^{-1})	carbon sequestration (Eur ha^{-1})	Erosion regulation (Eur ha^{-1})	Water regulation (Eur ha^{-1})	Invasion regulation (Eur ha^{-1})	Nitrogen removal (Eur ha^{-1})	Recreation (Eur ha^{-1})	Sum of values (Eur ha^{-1})
DG	Dry grasslands	370.72	17.22	265.48	1,875.90			55.45	2,584.76
AM	Alluvial meadows	864.09	68.88	265.48	2,755.00	9.80	161.95	55.45	4,180.64
MG	Mesic grasslands	695.39	43.05	265.48	2,113.00	52.00	161.95	55.45	3,224.37
WG	Seasonally wet and wet grasslands	883.55	68.88	265.48	3,042.00	20.41	161.95	55.45	4,497.71
AG	Alpine and subalpine grasslands	252.83	38.75	265.48	2,028.00	157.45		55.45	2,797.95
FF	Forest fringe vegetation	0.00	43.05	265.48	2,555.00			55.45	2,918.98
SM	Salt marshes	0.00	34.44	265.48	1,875.90			55.45	2,231.27
HT	Heathlands	0.00	25.83	265.48	1,268.00			55.45	1,614.76
P	Pastures and managed grasslands	403.71	43.05	265.48	1,639.00			55.45	2,406.69
	Average	385.59	42.57	265.48	2,127.98	26.63	53.98	55.45	2,647.96

Since the publication of the MEA report in 2005, there has been a very strong increase in the number of articles and communications on grasslands and ES, rising from less than 10 articles and communications per year before 2005 to 200 in 2015 (Web of Science consultation). It should be noted that the term 'ecosystem service' is quite recent and number of publications deal with the issue of the ES without naming it explicitly.

Most of the publications concern the work of ecologists, while agronomists or grassland scientists have published less on the topic. The main theme is biodiversity, either considering biodiversity for its role in the provision of ES (mainly the role of floristic diversity), or as a service provided by the ecosystem. In their review, Rodriguez-Ortega *et al.* (2014) found that the most evaluated ES of grasslands in the literature were (in descending order): biodiversity conservation, carbon sequestration, landscape quality, pollination, nutrient cycling, mitigation of soil erosion, flood control, and traditional ecological (cultural aspect). The UK National ecosystem assessment has highlighted that the main ES concerned by grasslands in the UK are the conservation of wild species diversity, landscape quality, pollination, soil and water quality.

According to the classification suggested by MEA, there is a large range of ES provided by grasslands (provisioning, regulating and cultural services) or for grasslands (supporting services). If we consider the European territory, there are very many types of grasslands, but the intensity of ES delivery hardly depends on the type of grasslands (Duru *et al.,* 2015b). Table 2 distinguishes the ES for semi-natural grasslands and agriculturally improved grasslands, following the classification suggested by Peeters *et al.* (2014).

Following the simple scoring matrices for ecosystem service production (Burkhard *et al.,* 2010), more advanced approaches to ecosystem service accounting are being developed. Ecosystem accounting is a relatively new and emerging field in statistics, dealing with integrating complex biophysical data, tracking changes in ecosystems and linking those changes to economic and other human activity.

Table 2. Grassland ecosystem services and their evaluation for semi-natural grasslands (SNG) and agriculturally improved grasslands (AIG).

	Ecosystem services	References	SNG[1]	AIG[1]
Provisioning	forage, milk, meat, fiber	Peeters, 2012; Taube *et al.*, 2014; Porqueddu *et al.*, 2016	0/+	++
	biomass for bioenergy	Wachendorf and Soussana, 2012; Murphy *et al.*, 2013; Herrmann *et al.*, 2014	0/+	++
	biomass for biorefinery	O'Keeffe *et al.*, 2011	+	+
	Medicinal and ornamental resources	Canter *et al.*, 2005	++	0
	wild flower germplasm for restoration and/or breeding	Rognli *et al.*, 2013; Haslgrubler *et al.*, 2014	++	0
	clean water supply	Keeler and Polasky, 2014	++	+
supporting	buffering/accelerating/slowing down of and nutrient cycling	Whitehead, 1995	++	++
	water infiltration and retention in soil	Ehlers & Goss, 2003	++	++
regulating	erosion control	van der Ploeg *et al.*, 1999	++	++
	flooding control	van der Ploeg *et al.*, 1999	++	+
	C-storage in soil	O'Mara, 2012	++	+
	greenhouse gas mitigation	Conant, 2010; Bellarby *et al.*, 2013; Jerome *et al.*, 2013	++	+/0
	wildfire control	Porqueddu *et al.*, 2016	+	++
	water purification	Eriksen *et al.*, 2010	++	+
	preserving biodiversity	Wrage *et al.*, 2011	++	+/0
cultural	grassland as herited cultural landscape	Quetier *et al.*, 2010	++	+
	leisure activities/recreation	Paletto *et al.*, 2015	0/+	0/+
	ecotourism	Harrison *et al.*, 2010	++	+

[1] 0 = no benefit, + = moderate benefit, ++ = strong benefit.

Grassland-based livestock systems hold the greatest potential to deliver public goods (non-excludable, non-rival) across European agricultural systems (Cooper *et al.*, 2009). Both the public goods and ES concepts place human benefits and societal demands at the core of their definition, and the importance of considering the needs and perceptions of society is therefore implicit. Many of ES that are public goods are ignored in classical evaluation frameworks because public goods do not have market price (nonmarket goods) and are often ignored in policy design, so farmers do not get the appropriate incentives to provide them. Therefore, there is a need to objectively evaluate the multiple private and public goods that grassland-based livestock systems deliver to the society, including their trade-offs and synergies, from different perspectives: biophysical, socio-cultural and economic (Rodríguez-Ortega *et al.*, 2014).

Due to the varying subjective perceptions and interest of stakeholders, the portfolio of outcomes (private or public goods) of grassland-based livestock systems that are relevant for people can vary across regions, socio-economic and policy contexts, and cultural backgrounds (Randall, 2002). In Table 3 we present a non-exhaustive overview of nonmarket functions and ES provided by these systems. It is worth mentioning that some public goods are intrinsically linked to grassland-based livestock farming systems. These are the maintenance of cultural landscapes, the preservation of biodiversity and the reduction of risk of forest wildfires, particularly in Mediterranean areas (Cooper *et al.*, 2009). In addition, it is important to notice that not all farms or farming systems that are located in grassland areas provide the same outcomes. The type and amount of private and public goods (or negative externalities) delivered by a particular farm or system of production will depend on the use of on-farm and off-farm resources, the land use, the degree of intensification (Bernués *et al.*, 2011), and ultimately on the specific agricultural practices implemented by the farmer (Duru *et al.*, 2015a,b).

Table 3. Examples of public goods (nonmarket functions and regulating/cultural ecosystem services) delivered by grassland-based livestock systems in Europe.

Nonmarket functions (main types)	**Ecosystem services**
OECD (2001)	Oteros-Rozas *et al.* (2014); Rodríguez-Ortega *et al.* (2014)
Agricultural landscape	*Regulating*
Cultural heritage	Tree regeneration
Environmental outputs	Biological control
Rural viability	Air purification
Agricultural employment	Habitat for species
Food security	Fire prevention
Animal welfare	Soil erosion control
Hall *et al.* (2004)	Connectivity and seed dispersal
Agrarian cultural heritage	Maintenance of soil fertility
New agricultural economy	Pollination
Traditional agricultural economy	Microclimate regulation
Environmental	Hydrological regulation
Rural leisure activities	Ditch maintenance
Cultural	*Cultural*
Van Huylenbroeck *et al.* (2007)	Tranquility/ relaxation
Economic functions (economic growth, market development)	Recreation
Social functions (dynamism of rural communities, quality of life, local knowledge,	Cultural identity
cultural heritage, history, rural lifestyle)	Recreational hunting
Environmental functions (biodiversity, climate change, desertification, water quality and	Scientific knowledge
availability, pollution)	Environmental education
Cooper *et al.* (2009)	Bullfighting events
Agricultural landscapes	Aesthetic value
Farmland biodiversity	Cultural exchange
Water quality	Spiritual value
Water availability	Local ecological knowledge
Soil functionality	Rural tourism
Climate stability (carbon storage)	
Air quality	
Resilience to flooding	
Resilience to fire	

Uncovering the values of ES that are important for human well-being requires diverse tools that can embrace their multidimensional character. The most widely used methods for socio-cultural and economic valuation applied to grassland-based livestock systems were reviewed by Rodríguez-Ortega *et al.* (2014). As monetary valuation techniques are dominant, the assessments of ES are often biased toward the information provided by markets. Therefore, it is advisable to combine the use of deliberative (qualitative) and quantitative techniques.

Studies on ecosystem services delivered by grasslands in Europe

As described previously, there is no clear consensus on the definition of ES, resulting in very different approaches for their assessment. The objective of this section is to show, from a few studies, the variety of approaches conducted over the years in Europe to assess the ES delivered by grasslands and grassland-based systems.

In the first part (Study 1), the question of the perception of grassland ES by different social categories will be addressed. Two European research programs aiming to develop common sets of indicators will be shortly presented, and analysed in the perspective of the need (or not) of local adaptations (Study 2). Studies 3a to 3c compare three contrasting geoclimatic and land use situations, in Nordic, Atlantic and continental conditions. We are not setting out to cover the full range of grassland situations in Europe. In fact the topic of ES indicators for grasslands is too recent, and very few studies were performed to draw general conclusions from a wide range of situations.

Study 1. Comparison of socio-cultural and economic value of grasslands in Mediterranean and Nordic Regions: assessment by various types of beneficiaries

In Europe, there is increasing pressure to shift the Common Agricultural Policy (CAP) toward the supply of public goods. For this to happen, accounting for agri-environmental indicators in order to quantify the impacts of agricultural practice on the environment, and integrating these indicators into policy design is key.

Two grassland areas in Mediterranean (Bernués *et al.*, 2014) and Nordic (Bernués *et al.*, 2015) regions were evaluated in study 1. The Mediterranean case was a mountain (430 to 2077 m.a.s.l.) protected area with wide variation of precipitation (600 to 1000 mm) and vegetation (49% shrub rangelands, 29% to dense forest, 7% open forest rangelands, 7% crops, 1% mountain summer grasslands). Grazing livestock is the main agricultural activity (32,651 meat sheep, 700 goats, 1,199 beef cattle and 259 mares). Grazing areas include private and communal land and some of the main habitats are 6,170 (Alpine and subalpine calcareous grasslands), 6,210 (Semi-natural dry grasslands Festuco-Brometalia) and 6,230 (Species-rich Nardus grasslands). The Nordic case was characterized by mountains (up to 1,800 m.a.s.l.) and fjords. The annual average temperature is 2.6 °C and precipitation varies between 700 and 2,000 mm. The natural vegetation in the valleys ascends from southern boreal and middle boreal zones with coniferous and deciduous forests to the northern boreal zone located near the climatic treeline. Cultivated land at the bottom of the valleys is very scarce; the rest is natural grasslands, fresh water, seawater, glaciers and bare rocks. Animals are grazed in the mountains between the northern boreal (approximately 900 m.a.s.l.) and alpine zones. The number of grazing animals was 2,290 meat sheep, 1,285 dairy goats and 49 dairy cows in 2012.

A combination of deliberative (focus groups and interviews with farmers and other local stakeholders) and survey-based stated-preference methods (choice modelling) was used to, first, identify the perceptions of farmers, other local stakeholders and urban citizens on the diverse functions of grassland-based livestock systems and, second, to value the corresponding ES in economic terms according to the willingness to pay (WTP) of the local (residents of the study areas) and general (regions where the study areas are located) populations.

In the Mediterranean mountains, the deliberative research with local farmers and urban residents showed many relationships between grassland-based farming and the environment; with greater importance for the aesthetic and recreational values of the landscape, the maintenance of biodiversity and the prevention of forest fires. The availability of high quality food products linked to the territory was also highly valued, especially by urban citizens. In the fjord and mountain regions of northern Europe, the functions considered as most important by the stakeholders were the control of forest growth, the maintenance of cultural heritage, the maintenance of rural life and activity, the conservation of soil fertility, the maintenance of tourism attraction, the maintenance of traditional agricultural landscape, the conservation of biodiversity, and the production of local quality foods.

In the second stage, a representative sample of the local and general populations were asked to choose their most preferred level of delivery of the most important ES above under three policy scenarios (see full details in Bernués *et al.* (2014) and Bernués *et al.* (2015)). The status quo scenario corresponded to the current delivery of ES, whereas the liberalization (reduction of agri-environmental support) and targeted support (additional funding to agri-environmental schemes) scenarios represented different combinations of levels of ES delivery (see example for Nordic grasslands in Figure 1).

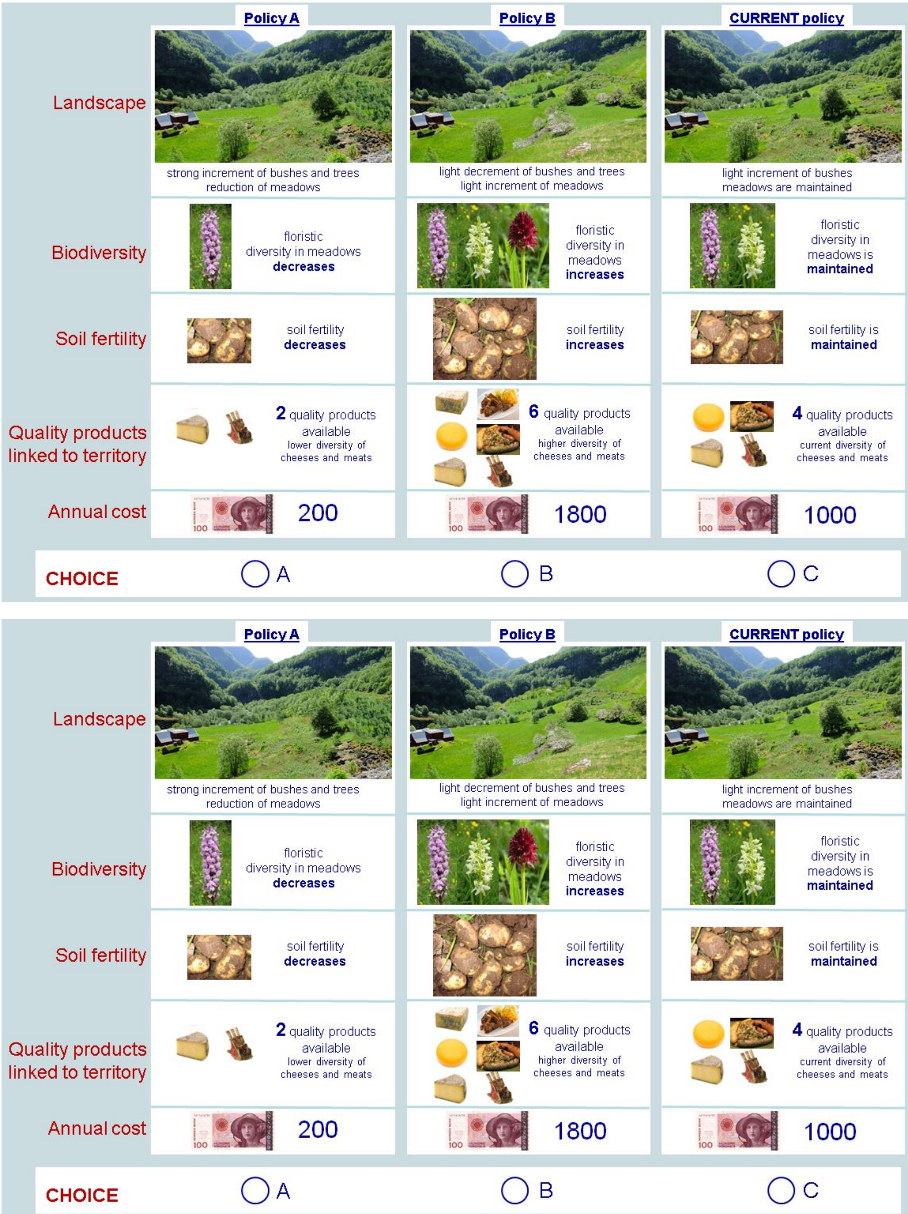

Figure 1. Choice set in Nordic grassland systems. The attributes (agricultural landscape, biodiversity, soil fertility, availability of quality products linked to the territory and societal cost in Norwegian kroner) presented here for illustration correspond to the liberalization (policy A) and targeted support (policy B) scenarios. Source: Bernués *et al.* (2015).

The analysis of responses with a mixed logit model allowed a ranking of ES according to the willingness to pay by society for their delivery (Table 4). In the Mediterranean, the prevention of forest fires was valued by the general population as a key ecosystem service delivered by mountain agroecosystems (≈50% of total WTP), followed by the production of specific quality products linked to the territory (≈20%), biodiversity (≈20%) and cultural landscapes (≈10%). For the local population, prevention of forest fires and production of quality products were also first in importance, however, the rank and importance of the cultural landscape and biodiversity varied (25% and ≈10 of WTP, respectively). The Total Economic Value of Mediterranean mountain agroecosystems was ≈120€ per person per year, three times the current level of support of agro-environmental schemes.

In Nordic regions, for the general population all ES had similar importance. The production of quality products (≈28% of total WTP) was valued as a key ecosystem service, followed by the preservation of soil fertility (≈27%), the conservation of the agricultural landscape (≈23%) and the conservation of biodiversity (≈22%). The values given by local residents were rather different; agricultural landscape represented 36% of total WTP, followed by the production of local quality products (≈33%) and soil fertility (≈19%), whereas biodiversity was valued rather low (≈11%). The total economic value of Nordic fjord and mountain agroecosystems was ≈850€ per person per year, seven times the current level of support of agro-environmental schemes. WTP of the local sample in the Nordic region could not be estimated as the parameter for cost was positive (see Bernués *et al.,* 2015 for explanation).

Large differences in the WTP were observed between regions. In Nordic countries the WTP was much higher than in the Mediterranean, both in absolute terms and in relation to the level of public expenditure on agro-environmental measures. This could be due to differences in the level of wealth between regions, but also to the fact that open farmland is very scarce in Nordic countries and forest is very predominant, so people are more sensitive to further abandonment of farming and loss of agricultural landscapes. For the ranking of importance of ES, however, the differences were smoother between regions. Use values, either direct (product quality) or indirect (forest fires and soil fertility in Mediterranean and Nordic regions, respectively), were ranked higher in general. The general population in Nordic regions gave similar importance to all attributes, with higher scores for product quality and soil fertility. However, the general population in the Mediterranean gave more than 50% of WTP to forest fires. In general,

Table 4. Willingness to pay (WTP) (€ person^{-1} year^{-1}), composition of the total economic value (TEV) (%), and rank of ecosystem services. (Bernués *et al.*, 2014, 2015)

		General sample			Local sample		
ES	**Value component of TEV**	**WTP**	**%**	**Rank**	**WTP**	**%**	**Rank**
Mediterranean							
Landscape	Non-extractive direct use	10.0	8.2	4	49.5	25.2	3
Biodiversity	Non-use existence	22.2	18.3	3	17.4	8.8	4
Forest fires	Indirect use	64.4	53.2	1	79.3	40.3	1
Product quality	Extractive direct use	24.5	20.2	2	50.6	25.7	2
	TEV	121.2	100.0		196.8	100.0	
Nordic							
Landscape	Non-extractive direct use	196.3	23.1	3	–	36.1	1
Biodiversity	Non-use existence	190.1	22.4	4	–	11.1	4
Soil fertility	Indirect use	228.9	26.9	2	–	19.3	3
Product quality	Extractive direct use	235.0	27.6	1	–	33.5	2
	TEV	850.3	100.0		–	100.0	

agricultural landscape (with the exception of the Nordic local population) and biodiversity (especially in local populations in both regions) were ranked comparatively low.

From the results of this study we can derive that there is a clear underestimation of the socio-cultural and economic values of ES provided by European grasslands. Furthermore, the social welfare loss linked to further abandonment of livestock farming in these areas and the environmental degradation associated to a scenario of policy liberalization is very large. From a societal perspective, it is therefore necessary to jointly measure, and include in policy design, the biophysical, socio-cultural and monetary values of ES (market and nonmarket) provided by grassland agro-ecosystems in order to promote their sustainability.

Study 2. European common set of indicators developed in European research projects related to grasslands ecosystem services: Multisward and Biobio

The question of the development of indicators to assess the ES delivery of grasslands lies between two opposing positions. On one side, the evaluation of a wide diversity of grasslands and of biogeographic regions requires the development of specific indicator systems able to deal with the contrasts involved in the compared situations (Van Ittersum *et al.*, 2008). On the other side, because the bio-geographic, pedo-climatic and socio-economic conditions vary widely throughout Europe (Huyghe *et al.*, 2014), indicator systems allowing the evaluation of strongly contrasting situations are necessary for pan-European assessments for the adoption of common agri-environmental measures, or to share a common view (like in the Mapping of Ecosystems and Ecosystem Services European initiative). Two recent FP7 European research projects (Multisward and Bibio) clearly opted for the second position, while trying to cover a wide variety of situations.

MultiSward

During the FP7 EU project MultiSward (multi-species swards and multi-scale strategies for multifunctional grassland-base ruminant production systems), two types of indicators were developed or used, according to three system levels: at field, at farm type or at regional level.

At field level, a biodiversity indicator system sensitive to management and to the characteristics of the grassland plant community was developed and partially validated (Plantureux *et al.*, 2014). This indicator system estimates the abundance and species richness of bumblebees, spiders and earthworms (also butterflies and grasshoppers) at the field level based on agricultural management and the botanical composition of the grassland. Therefore, these indicators can be considered as 'pressure' indicators (assessing the effects of practices on biodiversity) and as 'state' or impact' indicators (predicting potential biodiversity status for various taxa). Data on agricultural management and botanical composition should thus be available from field surveys to calculate these biodiversity indicators. In order to set the system of indicators, knowledge about the factors determining total species richness and abundance was derived from scientific literature, available database and expertise. No distinction was made between specific biogeographic zones, in order to apply these indicators very widely in Europe. The validation of the indicator resulted in controversial results. A positive correlation was always found between indicator values and observed species richness and abundance of all taxa. Nevertheless, the precision of the prediction was always quite low, due to the impossibility of finding studies covering the whole of the European conditions for all taxa, and the comparable data set for the validation of indicators.

At regional level, an indicator system was developed to evaluate the impacts of grassland-based ruminant production systems on air, soil and water quality, energy use and biodiversity. This indicator system was constructed to allow assessments at the region level across a wide range of conditions in Europe, as well as at the farm type level. Its structure is based on the DPSIR framework of the European Environment Agency (EEA, 1999). The 28 agro-environmental indicators of the European Commission (European

Commission 2006) formed an important basis for the indicator selection, but the MultiSward indicator system focuses on the provisioning and regulating services of grassland-based ruminant production systems and its scope is thus more restricted than the one of the agro-environmental indicators of the European Commission. A set of 45 indicators on the effects of agricultural land use was selected for the assessment at the regional level. With regard to biodiversity, the selected indicators were: (1) proportion of High Nature Value grasslands in the agricultural area; (2) number of Natura 2000 grassland habitats; (3) proportion of Natura 200 grassland habitats in the agricultural area; (4) Shannon diversity and equitability indices of land use type; (5) population trend of farmland birds; and (6) Shannon diversity and equitability indices of grazing livestock species (domestic diversity).

For the assessment at the farm type level, a set of 21 indicators were selected, most of them being also included in set of indicators for the region level. This difference between the two sets of indicators was necessary, because some of the data required are only available at the region or at the farm type level. The availability of data is a widespread difficulty often preventing the calculation of indicators. In MultiSward, the availability of data within institutions of the European Commission was thus an important criterion for indicator selection.

The indicator-based assessment at the regional level was implemented on 12 contrasting NUTS 2 regions (NUTS for Nomenclature of Territorial Units for Statistics), which were first classified in the following typology of livestock regions based on Pflimlin *et al.* (2005): (1) lowlands of temperate Europe; (2) grassland regions; (3) grassland and maize regions; (4) forage crops regions with temporary grasslands and maize; (5) arable land and livestock regions; (6) arable land and no livestock; (7) wet mountain regions; and (8) mediterranean regions.

The assessment at the region level included all farm types within each region. The indicator-based assessment was also implemented on 7 farm types across the 12 regions mentioned above. The farm types were defined according to the farm type approach of the European Commission (European Commission, 2013): (1) specialists field crops; (2) specialists dairying; (3) specialists cattle rearing and fattening; (4) cattle-dairying, rearing and fattening combined; (5) sheep, goats and other grazing livestock; (6) mixed livestock, mainly grazing livestock; and (7) mixed crops-livestock.

The results revealed large differences in the contribution of agro-environmental payments to the farm income between regions, as well as large differences in the evolution of the area under permanent grassland: the area under permanent grassland remained quite stable between 2000 and 2010 in the Grassland regions, while it decreased in the Grassland and maize regions and in the Arable land and livestock regions. Differences were also found between farm types, the proportion of permanent grassland in the agricultural area being markedly larger in the specialist cattle-rearing and fattening type than in the specialist dairying type. With respect to biodiversity conservation, the authors concluded that both the proportion of High Nature Value farmland and of Natura 2000 habitats in the agricultural area mainly reflect national policies. The analysis of the population of farmland birds indicated that the declining trend could not be stopped yet in all studied regions.

The use of data available in the statistical databanks of the European Commission make pan-European assessments financially conceivable, but the reliability of such indicators should still be validated by comparing the results obtained with such indicator systems with those from field measurements.

BioBio

The main objective of the FP7 EU project BioBio (Indicators for Biodiversity in Organic and Low Input Farming Systems) was to develop a biodiversity indicator system applicable at the European scale. Biobio was not focused on grasslands, but this project illustrates very well the development of indicators in relation to ecoclimatic region and land-use systems. To achieve this objective, a first set of indicators was selected based on a large literature review and a first stakeholder consultation (Herzog *et al.*, 2012). Twelve studies across Europe were then used to field-test the indicators and keep the most pertinent of them to evaluate habitat and species diversity, as well as the genetic diversity of domestic animals and crops. Beside direct indicators for habitat, species and genetic diversity, farm management metrics were kept as indirect indicators. The species richness of four taxonomic groups representing different trophic levels were selected as proxy for general species diversity: (1) vascular plants; (2) wild bees and bumblebees; (3) spiders; and (4) earthworms. In the BioBio indicator system, the species richness of these four taxonomic groups and habitat diversity has to be assessed in the field. About 15 person days per farm are necessary to assess the complete set of selected indicators (Herzog *et al.*, 2012). The species diversity data collected during the project were used for a pan-European assessment of organic farming on biodiversity (Schneider *et al.*, 2014) and Herzog *et al.* (2012) propose to use the BioBio indicator system as basis for the development of a biodiversity monitoring scheme for European farms. Data collection on a large number of farms would be expensive but the implementation of such an indicator systems would provide detail information on the state of biodiversity on agricultural land. The limits of such a system should nevertheless be fully considered when interpreting the results. Indeed, indices of species richness allow qualifying the ecosystems with respect to biodiversity, but these indices do allow ranking the ecosystems for their conservation value across bio-geographic regions because of the existing influence of the bio-geographic region on biodiversity. Moreover, habitat diversity was assessed using a range of habitats, but because the system was developed to compare a wide range of agricultural productions (included horticulture and vineyards) and was not focusing on grassland-based production, grassland types were only segregated in either 'grasslands managed for the primary purpose of agricultural production' or semi-natural habitats. This would be insufficient to differentiate habitat and species diversity at the farm level between grassland-based farming systems, because such systems often manage many contrasting grassland habitats (Duru *et al.*, 2013).

Combining the BioBio and the MultiSward approach might allow improving the comparison of grassland-based farms without adding to the work load necessary to implement the BioBio system. Such a combined approach remained to be tested.

Studies 3a, 3b and 3c. Ecosystem services delivered by grasslands in specific conditions

Scandinavian grasslands

In large parts of Scandinavia, grasslands still play an important role in ruminant based livestock production. In Norway, grassland and ruminant based production is the dominant form of agricultural production along the coast in western and northern Norway as well as in the mountain region in central Norway. Grasslands are thus important for ES related to food production in these regions. The regions dominated by grassland based food production are also important regions for tourism including the fjords and several on-shore archipelagos in the north. The importance of grassland-based agriculture as landscape management is therefore large although often hard to quantify. ES related to landscape aesthetics and recreation but also cultural values/heritage are therefore important.

The abandonment of marginal agricultural land in these regions is a threat to landscape, biodiversity and potential for agricultural production (Sang *et al.,* 2014; Wehn, personal communication). Land that is abandoned over time turns into forest through succession. This changes the character of the landscape which in turn may have large impact on tourism and recreational use of the landscape (Bryn *et al.,* 2013). Although large parts of Norway below the forest line are forest and only 3% of the land is agricultural land the openness of agricultural landscape is conceived as attractive and valuable (Daugstad, 2008, Ode

Sang and Tveit, 2013). The open grasslands, and especially the semi-natural grasslands, are important for biodiversity (Johansen *et al.,* 2015). In the national monitoring program for ecosystems and biodiversity, an index has been developed specifically for open semi-natural habitats in the lowlands (Johansen *et al.,* 2015). The index is composite index including 29 indicators related to grasslands and coastal heathlands. This includes both indicators based on individual species but also an indicator of successional status in semi-natural grasslands that are based on expert assessment. The later expert based indicator contains data on all 428 municipalities in Norway and is given more weight than species based indicators in the overall index (Pedersen *et al.,* 2016). The effects of successional change on populations can for some species be delayed by a time-lag in the response e.g. due to the longevity and clonal growth of many plant species in grasslands (Johansen *et al.,* 2015). Indicators involving expert assessments can to some extent take this into account but it should nevertheless be a long term objective to increase the number of data-based indicators of biodiversity and ES. In Sweden, aerial photos have been used extensively for monitoring changes in grasslands and landscapes more general through the National Inventory of Landscapes in Sweden (NILS) (Ståhl *et al.,* 2011).

An ES related to grasslands that has attracted quite some attention in the Scandinavian countries is carbon sequestration. The impact of climate caused by agriculture is considerable and something that has to be taken seriously by both governments and the industry. In Norway this discussion also involves questions related to livestock grazing in mountains and subalpine areas. The practice of grazing with free ranging livestock has a long history and has had large influence on landscape and vegetation in low-alpine and subalpine areas (Olsson *et al.,* 2000; Wehn *et al.,* 2012). Livestock grazing is a major factor for tree line dynamics (Hofgaard, 1997; Hofgaard *et al.,* 2010; Speed *et al.,* 2010) and the creation of patches with grasslands and grass- and herb-dominated subalpine forests (Wehn and Johansen, 2015). Reduced grazing by livestock in these areas will increase canopy cover in the subalpine forests and the tree line will gradually move upwards to higher altitude (Bryn *et al.,* 2013). The increase in forest cover and timber stock can contribute to increased carbon sequestration in biomass and soil (Goodale *et al.,* 2002). However, recent studies indicate that the story is a bit more complex. Increased forest cover in low-alpine and subalpine areas will influence snow cover dynamics during winter and early spring and this will in turn influence reflections of solar radiation, i.e. the albedo effect. It has been estimated that increase global warming due to reduced albedo can be as high as 10-17 times the effect of carbon sequestration due to increased forest cover (De Wit *et al.*, 2015). In Scandinavia, a better understanding of the relationship between grassland-forest dynamics and climate impact of grazing and agricultural land use is crucial and information and indicators from the lowland of other European countries cannot be used without thorough adaptation.

Machair grasslands in North-west Scotland

Machair is a distinctive type of coastal grassland, listed on Annex 1 of the EU Habitats Directive, which is found in the north and west of Scotland and in western Ireland. In a strict sense, 'machair' refers to a relatively flat and low lying sand plain formed by dry and wet (seasonally waterlogged) short-turf grasslands above impermeable bedrock, a habitat termed 'machair grassland'. It is estimated that 'machair grassland is restricted to about 25,000 ha in world-wide extent, with 17,500 ha in Scotland and the remainder in western Ireland (Maddock, 2008). Machair grasslands are complex features in terms of origin, development, processes, local habitat types and management. They are formed from sand blown inland following the periodic breakdown of foredunes above the beach and contain a mosaic of wet and dry grassland communities. These are related to grazing and tillage history superimposed upon gradients of surface stabilization, soil acidity, and salinity which are controlled by local sand blow, water-table fluctuation and micro-topography, giving rise to highly complex habitat mosaics (Lewis *et al.,* 2014b).

No plant sub-communities of the UK National Vegetation Classification are confined to machair, but the two most indicative are the *Festuca rubra-Galium verum* fixed dune grassland, *Ranunculus acris-Bellis perennis* sub-community of dry machair and the *Festuca rubra-Galium verum* grassland, *Prunella vulgaris* sub-community of wet machair. Few rare plant species are largely restricted to machair systems with an exception being the endemic orchid *Dactylorhiza majalis scotica* (Maddock, 2008). The machair environment is regarded as being important as one of the last areas in Britain supporting old field successions, some of which are a century or more old. The great complexity and diversity of habitats and plant communities within machair systems is also a special feature. Two rare bird species, corncrake *Crex crex* (which is globally threatened) and corn bunting *Miliaria calandra*, are associated with machair systems, and breeding wader populations on the machairs of the Uists, Tiree and Coll are regarded as the most important in the north-west Palaearctic (Calladine *et al.*, 2014).

Machair has a very long history of management by local communities over several millennia. In recent times this has involved a mix of seasonal extensive grazing (mainly by cattle, with pastures rested in the summer) and low-input low-output rotational cropping based on potatoes, oats and rye. This traditional mixed management sustains varied dune, fallow and arable weed communities which offer in some areas superb displays of flowering colour across wide expanses of unfenced land in summer (Long, 2009).

The flower-rich areas of the machair are important for a number of rare and declining insects such as the as the great yellow bumblebee *Bombus distinguendus*, which requires a succession of suitable forage species in order to successfully complete its cycle and reproduce (Charman *et al.*, 2009). The wider machair system also has a rich invertebrate fauna (McCracken, 2009).

Machair is a living, cultural landscape and much of its conservation value is dependent on the maintenance of viable crofting agriculture based on low-input shifting cultivation. Machair is highly susceptible to agricultural modification and is particularly sensitive to changes in grazing, sand and shingle extraction, and recreational impact (Maddock, 2008). Over recent decades here has been a substantial fall in the amount of arable cropping and a concentration of this land use practice on North and South Uist. Associated with this change is a reduction in strip cultivation, increased use of inorganic fertilisers and increased plough depths, all resulting in a decline in wildlife value of the machair system (Pakeman *et al.*, 2011; Lewis *et al.,* 2014).

To-date, studies of machair have primarily focused on the biodiversity ecosystem services associated with this type of grassland. As indicated above, these have largely been focused on the vegetation of the machair (as assessed by plant species composition and functional diversity of the species present) and the importance of this habitat for populations of breeding waders such as lapwing *Vanellus vanellus*, dunlin *Calidris alpina*, ringed plover *Charadrius hiaticula* and redshank *Tringa totanus*. There has, however, been little focus on other ecosystem services such as the contribution that machair grasslands make to agricultural production. Although there is general acceptance that machair provides good grazing for livestock and provides a good environment for outwintering livestock there has been no specific quantification of the provisioning services associated with machair. This seems surprising given that the continued existence of the habitat is so reliant on the continuation of low-intensity agricultural management practices. Although machair is specific to coastal areas of north-west Europe, there are many other grassland habitats across Europe which are intimately associated with what is now referred to as High Nature Value farming systems. A greater concentration on the wider ecosystem services associated with HNV farming systems is required in order to help justify increased financial support to these systems, since over 20 years if highlighting their importance from a purely biodiversity perspective has not been sufficient to prevent their continuing decline and abandonment, with adverse impacts on the habitats themselves.

Grasslands in Continental conditions (National study Czech Republic)

In a pilot study (Vačkář *et al.,* 2010), a habitat approach to ecosystem accounting was applied which is based on a classification of habitat types. Grassland habitat types are regarded as ecosystem assets which provide vital ecosystem services. Grassland ecosystems were defined as habitats dominated by grasses, herbs and sedges. Broader grassland natural habitat type categories were identified, spanning the continuum from wetlands to rock succulents. Habitat Mapping Programme coordinated by the Agency for Nature Conservation and Landscape Protection of the Czech Republic consistently mapped the area and quality of natural grassland habitats. Natural grassland habitats cover nearly 3,000 km^2 which is about 4% of the total territory of the Czech Republic. Permanent pastures and meadows cover 11.7% of total land area and 22.5% of utilized agricultural area of the Czech Republic. The Classification of habitat types of the Czech Republic was combined with EUNIS and Corine Land Cover classification to delineate 8 semi-natural grassland habitat categories

The study accounts for assessment of several ecosystem services which contribute extensively to the benefits provided by grasslands. These services, namely livestock provision, carbon sequestration, soil erosion regulation, water flow regulation, invasion resistance and recreation were further supplemented by waste treatment (i.e. nitrogen removal). This survey made the assessment of all these services complete by calculating both biophysical quantity and economic value of each service.

The results of the ecosystem services assessment provided by natural and managed grasslands and pastures in the Czech Republic indicate that grasslands provide valuable bundles of ecosystem services and benefits that are not accounted for in current systems.

The total value of grasslands in the Czech Republic based on ecosystem services assessed is 2,578 million EUR per year in 2010. The total value of services provided by managed pastures and meadows slightly exceeds the value of services provided by natural and semi-natural habitats. The value of the annual flow of services from the pastures reaches 1,429 million euros and the value of services of natural habitats 1,149 million euros. However, due to the smaller overall area of natural habitats, these habitats achieve much greater performance in the provision of ecosystem services, which is reflected in the estimated ecosystem cost 3,134 EUR per hectare of natural habitat and 2,035 EUR per hectare of managed grasslands. Natural and semi-natural habitats to achieve higher values especially in the case of regulation of surface water runoff, nitrogen removal and invasion control.

Highest value of ecosystem services was reached in seasonally wet and wet grasslands, followed by alluvial meadows. Both habitat types provide service values more than 4,000 EUR per hectare. These habitats are followed by mesic grasslands which still provide multiple ecosystem values. Forest fringe vegetation, alpine and subalpine grasslands and dry grasslands provide comparable benefits in the range 2,585-2,919 EUR per hectare of habitat. Pastures and managed meadows provide relatively low economic values compared with semi-natural grasslands. Only salt marshes and heathlands were found to provide lower economic values per habitat area. The dominant component of ecosystem services is water flow regulation, followed by livestock provision and erosion regulation.

The assessment and valuation of grassland ecosystem services contributes to the discussion on the effectiveness of agri-environmental schemes and landscape management programs.

Discussion and conclusions

The question of the selection of indicators of ecosystem services in relation to ecoclimatic regions and the land-use systems must finally take many aspects into account:

The first aspect is certainly that of the final goals of these indicators: For whom are these indicators intended and what for? According to the definition of Mitchell *et al.* (1995), an indicator provides information about a complex system to facilitate its understanding [...], so that the users of the indicator can make appropriate decisions leading to the achievement of the objectives. Four types of users of indicators and uses can be distinguished: (1) society in general to be informed of the level of provision of ES of agro-ecosystems, and if necessary be alerted to threats in the short or long term on these ES, (2) public authorities (Europe, States, regions, ...) which are in charge of set up and evaluate policies and in some countries of the protection of environment, (3) scientists or all those who study the functioning of agro-ecosystems and evaluate their performance, and (4) managers (in particular farmers) and their advisers to assess the positive or negative impacts of their actions, and provide corrective actions. All these categories do not necessarily need same indicators, e.g. society is primarily concerned with indicators assessing ES (processes or benefits) associated with these services, while farmers also deal with indicators of pressure on these ES. Generally speaking, the importance for society of the different ES (and perceptions of different stakeholders) might differ, this is why together with biophysical valuation, socio-cultural and economic approaches are relevant. Sometimes the indicator might be relevant for many stakeholders, but the explanation (and metrics we use) should be adapted to different audiences.

In relation to the first aspect arises the question of spatial and temporal scales, and organization levels (hereafter scale) at which these indicators are calculated. The studies provided as examples in this paper illustrate the variability of possible scales: field, farm, Region, State or Europe. The four types of identified users above have not necessarily expectations at all these scales. At very large scales (regions, State, Europe), the level of precision of local data is often low, and the objective to draw maps of provision of ecosystem services led to the selection of a small number of services, easily quantifiable from official statistical sources, and common to compare regions and statistical or countries between them. This is the case in what has been undertaken at the European level for the MAES (Mapping and Assessment of Ecosystems and their Services). In contrast, the more local studies of the ecosystem services provided by grassland may justify selecting more specific indicators like in the case of plant and bird species followed in the Machair grasslands in Scotland. In this case there is often indicators that are common to many ecoclimatic situations, but with methods of calculation adapted to the local context. For instance a common indicator of 'regulation of water quality' may be chosen, but the criteria for assessing the quality of water vary according issues of each situation (pesticides, nitrate, phosphate, acidity, oxygenation,..). Another point of view is to consider that a common indicator would be an indicator considering the same criteria in all situations but weighing them differently according to the issues of each situation. The question of time scales is also of interest, as some ES operate in the short term (e.g. provision of forage) while others operate at longer scales (e.g. landscape). Also, from the farm perspective, ecosystem disservices (or impacts) are easy to quantify at a precise time (e.g. N pollution) but ES have much wider temporal and spatial scales.

Clearly there is no consensus on the definition of ES. Some propose to consider the conditions and processes of the ecosystem at the origin of benefits, whereas the MEA includes in the ES processes, benefits and market and public goods. In absence of agreement on this point, our recommendation is to give, before selecting indicators of ES, the precise definition of each ES, the reasons for its choice and the beneficiaries. For grasslands, using the term pollination is thus insufficient, and it should be indicated if the ES is that of pollination of grassland plants, or wild or cultivated plants in the edges or crop fields close to the grasslands, or even if one thinks of the maintenance of wild pollinators or honeybees. It is also important to be precise over the actual or potential beneficiaries: society, farmers, beekeepers, ... A recurring issue for the grasslands is how to consider the livestock: as an element of ecosystem that provide ES or as a biological entities that transform ES provided by grasslands. In other words, is forage production or the production of milk or meat an ES of grassland? We previously stated that the portion of goods originating from the anthropogenic capital should not be consider as ES and that only the portion of goods supported by the ecosystem without anthropogenic capital should be consider as ES. In the case of grassland, the portion of forage obtained with fertilizer should thus not be considered as ES. This issue of the definition of terms is not just a semantic problem, but rather a scientific and a political question.

The vision of the ES provided by grasslands is still often a static vision, although long-term and dynamic assessments are frequently needed. At best, tracking over time the evolution of an ES is to provide information on the favourable or unfavourable evolutions of the provision of service. It would be important to also evaluate the resilience of the grasslands ES, and imagine related indicators, to face changes or disruptions to natural (climate, biological) or human (management, economy) environment. The ability of grassland to maintain a level of ES despite strong economic and climatic fluctuations is as important as the average level of service provided. The studies of grasslands and abandonment in Scandinavia that are mentioned above may provide examples on how ongoing land-use will influence ES from grasslands in the future. Still, these analyses need to be further developed by capitalizing on an increasing amount of relevant data and modelling tools. To do so, scientifically based and quantitative descriptions of the relationships between land-use and delivery of ES is needed and then preferably as production functions

Adaptation to local conditions (ecoclimatic and land-use) indicators raises two questions: (1) that of the relevance of the indicators for the ecoclimatic and/or land-use conditions, and (2) the availability of the data. On the first point, it may be interesting to make sure that the indicator is really suitable to the situation. Considering protection against water erosion in a flat region or degradation of pesticides in high mountains meadows is certainly irrelevant. Showing that a service is not provided by grasslands although it could be is much more interesting, for example the absence of maintenance of floristic biodiversity in very intensified prairies. The availability of data is a major problem for the calculation and the adjustment of indicators of SE. We must recognize that the number of ES indicators we are able to calculate accurately, reliably and systematically for grasslands, is still very low. Many ES (carbon sequestration, supply of nitrogen, ...) are estimated from models, and with simplifying assumptions. Even when the models are accurate enough, it often lacks reliable input data. The surveys and systematic observations on large territories are very costly in time and money. As noticed by Honigova *et al.* (2012), 'Although we did not find any study, which would comprehensively quantify grassland ecosystem services, the value of grassland ecosystems has been already addresses and assessed by several studies'. An interesting perspective comes from the emergence of new sources of information: acquisition of satellite images, creation of web databases and 'bigdata', acquisition of information by the society (farmers, public, ...). This could ultimately facilitate the calculation on a large scale of ES indicators. Economists of ES use the concept of benefit (or value) transfer methods for scaling up values. They extrapolate the value of an ecosystem to a similar one. This should be an interesting track for the reflection in biophysical terms with the aid of new information databases.

In conclusion, we think that it is necessary to develop indicators for ecosystem services adapted to ecoclimatic and land-use conditions. The issue of ES is a global issue, and thus it appeared desirable to find common indicators in order to compare the systems instead. In study 1 in Mediterranean conditions, it was demonstrated that the public normally gives attention of similar issues, like landscape, biodiversity, pollution, and one key particular aspect: the differential quality of the products coming from grassland farming systems. The key question should be: 'The question is: can we find indicators adapted to a wide range of ecoclimatic and land-use conditions that are still precise enough to compare systems within ecoclimatic and land-use conditions? This can be very important in the debate on the role and the maintenance of grasslands in livestock production systems. The economic competitiveness of grasslands is often questioned in favour of other forage resources. To be able to accurately assess all the ES delivered by grasslands, and compare it to the alternatives of production appears a major challenge for agriculture and the environment. There is a shared opinion in agriculture that that most of grasslands especially semi-natural ones are less efficient in terms of dry matter grass yield, but it very much depends on what efficiency is considered. The multifunctional character of grasslands and grassland based animal production needs to be upgraded, and assessment of ES could help in this perspective.

There are numerous types of grasslands in various conditions: for each combination, the trade-offs between ES are not the same, and strategies to optimize ES at local and global level needs to take in account this feature of grasslands. It is clear that, beyond the questions of definitions of ES, there is still a lot of scientific and technical knowledge to produce for the goal of an accurate and systematic assessment of the ES provided by grasslands.

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Linkages between biodiversity and ecosystem services in grazed and abandoned semi-natural grasslands?

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Abstract

In this study potential links between biodiversity, ecosystem service (ES) indicators and agricultural land use were examined. Semi-natural grasslands, either actively used for sheep grazing or abandoned, were surveyed and all vascular plant species recorded. Based on this survey, species richness (per 4 m^2), fodder quality and quantity, as well as pollination, were estimated. We found lower species richness and indications of lower fodder quantity and quality, but of higher pollination in abandoned grasslands. The relationships between ES indicators and species richness were both negative and positive, and differed in managed and abandoned semi-natural grasslands.

Keywords: provisioning services, land use, semi-natural grassland, abandonment

Introduction

Ecosystem services (ES) are fundamental for human well-being (MEA, 2005). Biodiversity is the foundation of ecosystem functioning and the worldwide decline in biodiversity therefore causes concerns about future provisioning of ES. Land use change is one of the greatest threats to biodiversity (Newbold, 2015; Norderhaug and Johansen, 2010). However, the link between biodiversity and ecosystem functioning is complex and the understanding of how changes in land use will impact ES is insufficient (Mayfield *et al.*, 2010). In agricultural landscapes, semi-natural grasslands typically contain a larger part of the biodiversity than expected from their area alone (Billeter *et al.,* 2008), but changes in agricultural land use have resulted in a loss of and altered species compositions in semi-natural grasslands (Norderhaug and Johansen, 2010). In this paper we focus on the effects of abandoned pastoralism on species richness and indicators of agricultural value in semi-natural grasslands. We also aim to assess how ES indicators are related to species richness. ES can be difficult to quantify directly due to costs and practical considerations, but characteristics of the ecosystem such as functional identity and diversity of plant communities can be used as ES indicators (De Bello *et al.*, 2010; Pakeman, 2014). In our study we used community weighted mean of (CWM) specific leaf area (SLA), leaf dry matter content (LDMC), and leaf nitrogen content (LNC), and abundance of (ABUN) graminoids as indicators for the quantity and quality of fodder produced in semi-natural grasslands. Furthermore, abundance of plant species attractive for *Hymenopterans* (ABUN *Hymenoptera*) were used as a proxy to measure the availability of feeding resources for pollinators in the vegetation.

Materials and methods

Study sites (n_{sites}: 14) were established in Western and Mid-Norway; each including one plot in a managed semi-natural grassland in an enclosure where sheep were allowed to graze in spring and/or autumn, and one plot in an abandoned semi-natural grassland. Four subplots (4 m^2; $n_{subplots}$: 110) were randomly located within each plot. Abundance of all vascular plant species was registered. Information on functional trait values/categories of the species were extracted from databases: LEDA (Kleyer *et al.*, 2008), TRY (Kattge *et al.*, 2011), and the Biological Records Centre's database of insects and their food plants. ABUN and CWM of the traits were calculated for each subplot, CWM by using the R-package FD version 1.0-12.

Likelihood ratio Chi-square tests were used to compare Linear Mixed Models. Both simple models and models including interactions between species number and abandonment were modelled by using the lme4 package in the R 3.1.1 software (R Core Team, 2015).

Results and discussion

Plant species number was highest in the managed semi-natural grasslands (P<0.001; Figure 1) indicating that grazing is important to maintain biodiversity in semi-natural grasslands, as was also pointed out by Metera *et al.* (2010). ABUN graminoids were higher in the managed grasslands (Table 1). CWM LDMC was lower, but as high values of LDMC indicate lower quantity of fodder (Pakeman, 2014), these two results both indicate higher agricultural value in grazed areas. By contrast, ABUN *Hymenoptera* was higher in the abandoned semi-natural grasslands.

Biodiversity is both negatively and positively related with provisioning of ES (Harrison *et al.*, 2014), as shown in our study (Table 1, richness relationship). ABUN graminoids were positively correlated with species richness, CWM LNC negatively, and finally CWM SLA, CWM LDMC, and ABUN *Hymenoptera* not. Further, for two of the indicators investigated (ABUN graminoids, ABUN *Hymenoptera*), the link differed in managed and abandoned semi-natural grasslands (Table 1 interaction; Figure 1); land use does influence the links between biodiversity and ES.

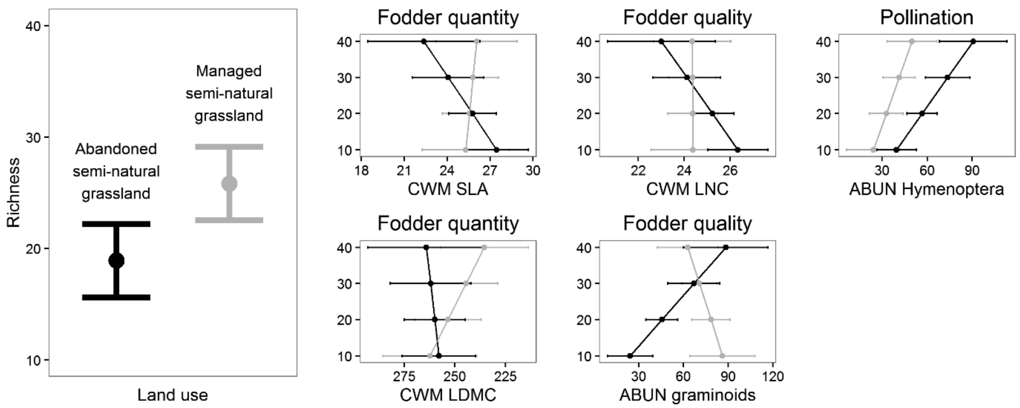

Figure 1. The estimated values and the confidence intervals of richness (number of plant species) and agricultural value indicators in abandoned (black) and managed (grey) semi-natural grasslands and the links between the indicators and plant species richness in the two land use categories.

Table 1. Effects of grazing abandonment on ecosystem service (ES; agricultural provision) indicators, relationships between ES indicators and plant species richness, and whether the effects of land use on the ES indicators vary along the richness gradient (interaction).

	Indicator[1]	**Abandonment**		**Richness**		**Interaction**
		effect	***P*-value**	**relationship**	***P*-value**	
Fodder quantity	CWM SLA	no	0.770	no	0.234	no
Fodder quantity	CWM LDMC	increase	0.048	no	0.105	no
Fodder quality	CWM LNC	no	0.110	negative	0.041	no
Fodder quality	ABUN graminoids	decrease	<0.001	positive	<0.001	yes
Pollination	ABUN *Hymenoptera*	increase	0.001	no	0.116	yes

[1] CWM = community weighted mean; SLA = specific leaf area; LDMC = leaf dry matter content; LNC = leaf nitrogen content; ABUN = abundance of graminoids/*Hymenoptera*.

Conclusions

Ceased grazing imposed negative influence on plant species richness and some, but not all, ES indicators. Plant richness was often positively correlated with ES indicators, but not always and different links between biodiversity and ES are present in different land use categories.

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Effects of grazed plant groups in Norwegian alpine rangelands on milk production and quality parameters

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Abstract

The main aim of this study was to identify potential relationships between chemical compounds (protein, fat, urea) in milk from dairy cattle, milk yield and distinct plant groups in the diet when grazing on alpine rangelands. Two herds in two different summer farming areas were studied during three grazing seasons. The results of statistical analyses indicate that milk yield was significantly related to different plant groups in the diet (herbs, grasses, sedges, woody species, Pteridophytes), but only herbs were positively related to yield. The composition of the different plant groups in the diet reflected the quality of the available vegetation types for grazing. The vegetation types were in various phases of scrub encroachment and forest regrowth. When available, dairy cattle preferred open areas with a vegetation rich in species, dominated by various herb and grass species.

Keywords: alpine rangelands, biodiversity, milk production

Introduction

Alpine summer farming with production of milk and dairy products on alpine rangelands was earlier a common practice in Norway. The grazing resources in the mountains were also utilised by goats, sheep and horses, and firewood and wild grazing plants were harvested for fuel and winter feed. Over the last decades, the grazing pressure on alpine rangelands has decreased, the harvest of firewood and winter feed has ended and semi-natural grasslands in the mountains have changed from being grass- and herb-dominated towards vegetation dominated by bushes and trees (Bryn *et al.*, 2001). Today, much of our production of milk is based on imported feed resources. The sustainability of this production and the possibilities for increased use of our own feed resources are, however, widely debated. Furthermore, the study 'Effects of vegetation and grazing preferences on the quality of alpine dairy products' (Sickel, 2014) concluded that milk from alpine rangelands may give rise to unique milk products because of its healthier fatty acid composition and its relatively high number of terpenes. Chemical components in the milk (various fatty acids and β-carotene) were related to various plant groups in the diet. In this study we have analysed data from that study on milk yield, fat, protein and urea levels in the milk, to investigate whether these parameters may also be related to the grazed plant groups.

Materials and methods

The study sites were two summer farms situated in south-central Norway, with dairy cattle grazing alpine ranges. The sites are referred to as site 1 (Valdres, 60°57 ′N; 8°49 ′E) and 2 (Hallingdal, 60°32 ′N; 8°11 ′E). Site 1 is situated 910 m a.s.l. in the northern boreal vegetation zone. Site 2 is located 1,040 m a.s.l. in the transition between the northern boreal and low alpine vegetation zone. The animals in both herds were of the breed Norwegian Red, and the size of the herds at site 1 and 2 were 12-14 and 18-20 dairy cows, respectively. The herds spent 9-11 hours per day on alpine rangeland. After the afternoon milking, the cattle were let out in fenced cultivated pastures where they spent the night resting and grazing until the morning milking. The proportion of total feed ration ingested on alpine rangeland was not measured. Observations of animal behaviour suggest that 60-70% of total feed intake from grazing was from alpine rangelands and 30-40% from cultivated pastures.

Five cows in each herd were equipped with a global positioning system (GPS). Area use and grazing preferences were studied by using high frequent GPS data in combination with detailed vegetation mapping, field studies of animal behaviour, microhistological analysis of faeces, analyses of rangeland vegetation composition and records of grazed plant species. For details about GPS tracking of the animals, development of GPS models that separate GPS positions in grazing, resting and walking behaviour, vegetation maps and GIS analyses, see Sickel (2014).

In all study years (2007-2009), milk samples (morning milk) were taken individually from the GPS cows in each herd, once in July and once in August at both study sites. The milk samples were subject to standard quality analysis of the content of fat, protein and urea levels by authorized laboratories in TINE Dairy Company. Data regarding milk amounts (kg), supplemental feeding with concentrates (type and amounts), age and calving date for each cow individually at the time of milk sampling were supplied by the TINE Dairy Company. Supplemental feeding consisted of grain based concentrates at both sites. The average amounts of fed concentrates were 5.2±0.9 kg day^{-1} at site 1 and 5.9±1.5 kg day^{-1} at site 2. The GPS cows at site 1 milked on average 15.8±3.9 kg day^{-1} while the GPS cows at site 2 milked 22.7±5.0 kg day^{-1}. The average age of the GPS cows was 3.7 years at site 1 and 3.8 years at site 2. The average months since calving were 7.0 at site 1 and 5.1 at site 2.

A set of individual diet variables was made from the results of the microhistological analyses of faeces samples taken from the GPS cows each day, 7 to 2 days before milk sampling. The species and plant families detected were grouped in five functional groups: woody species, grasses, sedges, herbs and Pteridophytes, and the proportions of grazed plant groups were calculated and used as individual diet variables: 'wd' (woody species), 'gd' (grasses), 'sd' (sedges), 'hd' (herbs) and 'pd' (Pteridophytes). Grazed plant species were also recorded by plot analysis (0.5×0.5 m) on grazed patches within the vegetation types. All species within the plot were recorded and scored with respect to whether they were grazed on or not. The proportions of grazed plant groups within each grazed vegetation type were calculated. For every GPS cow, a set of diet variables was calculated by multiplying the proportions of grazed plant groups within the vegetation type with the proportion of time spent grazing in the vegetation type the week before milk sampling, summing up for all grazed vegetation types and calculating the overall proportions of eaten plant groups from that week: 'we' (woody species), 'ge' (grasses), 'se' (sedges), 'he' (herbs) and 'pe' (Pteridophytes). For more details, see Sickel (2014).

To investigate potential relationships between diet, milk yield and the chemical composition of the milk, linear mixed modelling was performed using the milk data as response variables and different measures of cow diet, year (random term), month (fixed term), and the covariates age, concentrates (conc) and months since last calving (msc) as explanatory variables. The analyses were performed with proc mixed in SAS 9.4 and the linear mixed models were of the form: $y = intercept + year + month + age + conc + msc + x + error$, where x is a diet measure. Significance level was set to 0.05.

Results and discussion

Milk yield was found to be significantly associated with seven of the cow diet variables, and to 'age', 'conc' and 'msc'. It was positively related to 'hd' and 'he' and to 'age' and 'conc'. It was negatively related to 'pd' and 'pe' and to 'msc'. There were also negative relationships between milk yield and 'we', 'ge' and 'se'. Fat was positively related to 'hd' and 'age'. There were no significant relationships between protein and the diet variables or urea and the diet variables. Protein was negatively related to 'age' and positively related to 'msc'.

Herbs in the diet were the only plant group variables positively related to milk yield. There were significantly more herbs and sedges in the faeces from site 2 and more woody species and Pteridophytes in the faeces from site 1 (Sickel, 2014). GIS-analyses of GPS data and vegetation data showed that the mostly grazed vegetation types at site 1 were tall-herb mountain birch forest and bilberry woodland and that approximately 50% of the grazing time was spent there. The proportion of herbs grazed in the plots from these vegetation types were 17 and 10% respectively. Herb-rich vegetation types as intermediate grassland constituted only a small part of the grazing area at site 1, were mainly covered with 60-100% bushes and only 10% of the grazing time was spent there. At site 2 the most frequently grazed vegetation types were intermediate grassland with 0-59% bush cover and bilberry woodland (28 and 15% of the grazing time, respectively). The proportions of herbs grazed in the plots from intermediate grassland and bilberry woodland were 34-46% and 21%, respectively. Our interpretation of the positive relation between herbs and milk yield is therefore that herbs are an indicator of good grazing resources, with greater availability of herb and grass species with high forage quality. Together with slightly higher amounts of concentrates and an earlier state of lactation, this may explain the higher milk yield at site 2.

Conclusions

Results from this study may indicate that the composition of plant groups on alpine rangelands influences the production potential of milk from dairy cattle. The regrowth of semi-natural grasslands with bushes and trees results in vegetation types with fewer herbs and reduces the quality of the grazing resources. How we manage our alpine rangelands may in other words be essential for the future production potential of milk and meat in these areas.

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A French classification of permanent grasslands at national level to evaluate their forage and environmental services

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Abstract

To demonstrate and quantify the forage and environmental services provided by permanent grasslands, a research program (2008-2011) was performed, including together INRA, Institut of Elevage and 23 extension services. A national network of 190 permanent grasslands was selected from a database of 1,500 grasslands, to represent the diversity of management and agro-climatic conditions, and has been studied for two years. On each plot, a botanical survey was carried out (2009). Grass production, functional composition of the grassland and forage nutritive value for ruminants were measured on four dates during the season of growth and during two years observations (2009 and 2010). Data of functional and botanical composition were analysed to define 19 types of grassland identifiable from a simple decision key (agro-climatic conditions, management). For each type, quantitative references of seasonal evolution of functional composition, biomass production and nutritive value are provided. The contribution of the different types of grassland to ecosystem services was evaluated.

Keywords: permanent grassland, classification, production, environmental value

Introduction

The contribution of permanent grasslands in ruminant feeding is considerable and the economic interest of these surfaces in fodder systems is now recognized. Their environmental interest is also important, e.g. fauna flora biodiversity (Farruggia *et al.*, 2008) or carbon sequestration (Soussana and Lüscher, 2007). However, these surfaces have been decreasing in France for 50 years and are mainly replaced by maize for silage or more profitable annual crops. This decrease can be partly explained by a deficit of technical and scientific knowledge about the agronomic, ecological value and the management of the permanent grasslands. To revalue permanent grasslands in the forage systems, a R&D Program (funded by the French Ministry of Agriculture) was set up (2008-2011). The objective of this program was to build a reference tool (a classification) reporting the diversity of agronomic and environmental value and of some ecosystem services of French permanent grasslands. This tool was intended for education and actors of development with the aim of better managing and valuing of these surfaces in the fodder systems.

Materials and methods

A set of 190 permanent grasslands was selected from 1,500 listed by a survey conducted in 2008 on 78 farms (in which permanent grasslands represented more than 50% of forage area) distributed in the main lowland and mountain grassland areas of France except the Alps and the Mediterranean area. Information concerning management practices and the characteristics of the permanent grasslands of the network (location, type of soil) were listed. Botanical composition (exhaustive inventory of the species and the estimation of their dominance) was determined in spring 2009 on each grassland in a homogeneous plant community (Michaud *et al.,* 2011). The seasonal dynamics of forage production and nutritive value were assessed during 2009 and 2010 on the dominant homogeneous plant community of each grassland. The proportions of grasses, legumes and forbs were estimated visually, according to their volume, and on a sub-sample of 40 grass tillers, the proportions of functional types were determined according to Cruz *et al.* (2010). On dried samples, the nutritive value of the herbage (organic matter digestibility and crude protein content) was estimated using NIRS (Michaud *et al.,* 2015).

Principal component analysis based on six criteria of vegetation (proportion of legumes in the beginning and at the end of spring, proportion of forbs at the end of spring, proportion of functional type C in the beginning and at the end of spring (classification of vegetation according to nutriment strategy; Cruz *et al.,* 2010), proportion of function type B at the end of spring) contributed to build a classification of permanent grasslands. A first classification based on this criteria of vegetation and criteria of nutritive value allowed to identify 15 types of permanent grasslands related to forage services. At the same time, a second classification based on criteria of the vegetation and criteria relevant for the prediction of the environmental value (floristic composition and dominance of entomophilous species) contributed to identify 12 types of grassland related to environment services. Combining these two classifications resulted in classification into 19 defined types each with a particular combination of six characteristics of the vegetation retained. A key of determination was established, allowing us to recognize the types of grassland by leaning mainly on the nature of the environment (height, etc.) and management practices.

Results and discussion

The 19 types of permanent grassland are distributed in four groups according to the characteristics of climate and height: mountain permanent grasslands (5 types), plains and hills with semi-continental climate permanent grasslands (6 types), plains and hills oceanic permanent grasslands (5 types), Atlantic coast permanent grasslands (3 types).The proportion of legumes in biomass contributes to discriminate semi continental permanent grasslands. Management practices and particularly fertilization contribute to discriminate between all types of permanent grasslands.

Detailed descriptions of the 19 grassland types are given in the book by Launay *et al.* (2011). For each grassland type, an index card describes the bioclimatic conditions, soil type, management practices and the botanical characteristics (species present and dominant). Dry matter yield, nutritive value and specific richness are given as averages of two years (Figure 1). These values allowed us to estimate some ecosystem services (biomass production, biodiversity, pollination). This tool has the ambition of bringing references of agronomic values and ecosystem services provided by various types of permanent grassland.

Conclusions

This national classification reports the diversity of French permanent grasslands and so the diversity of ecosystem services provided. This classification of permanent grasslands constitutes a tool used in agronomic and environmental assessments. The methodology could be used in other European countries.

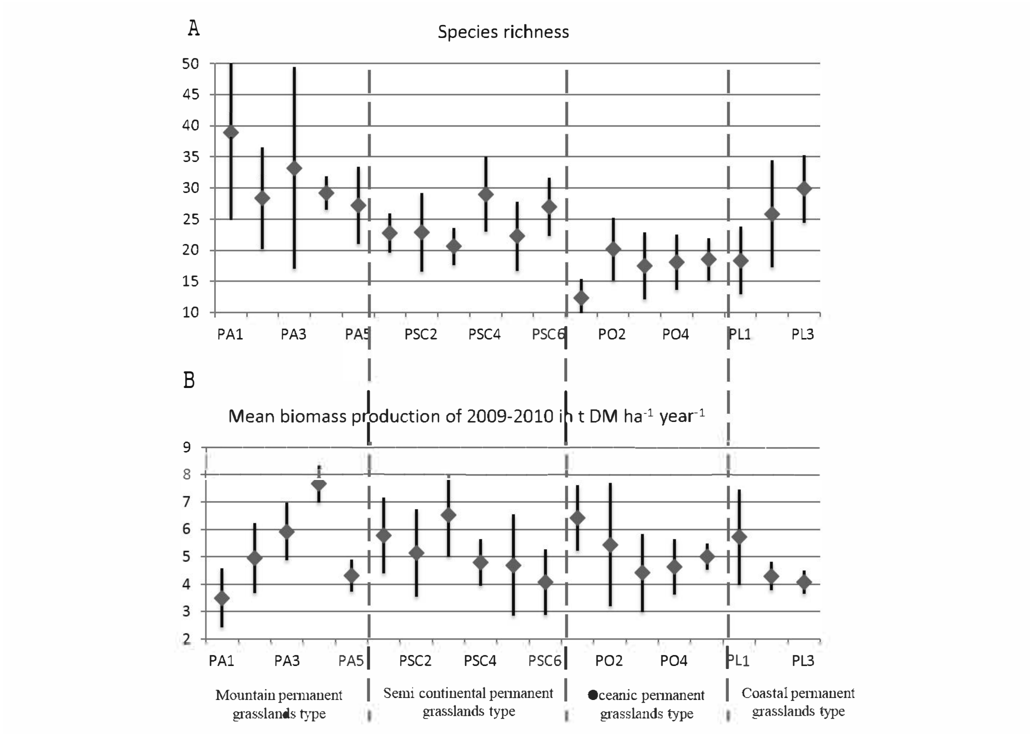

Figure 1. Example of references brought by the classification: (A) biomass production marks for each type of permanent grasslands in t DMY ha^{-1} year^{-1}), (B) specific richness in each type of permanent grasslands (number).

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Diversity promotes production of ryegrass-clover leys through inclusion of competitive forb species

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Abstract

Highly productive temporary grasslands in Europe are usually composed of only a few plant species, and are typically dominated by perennial ryegrass-clover mixtures. Including additional competitive forb species holds potential for enhancing productivity in temporary grasslands, but requires further demonstration. In a grassland biodiversity experiment, one or all of the three forb species: chicory (*Cichorium intybus* L.), caraway (*Carum carvi* L.) and plantain (*Plantago lanceolata* L.), were grown in different proportions with the perennial ryegrass (*Lolium perenne* L.)-red clover (*Trifolium pratense* L.) mixture under two slurry application levels (0 and 250 kg total N ha^{-1} year^{-1}). Dry matter (DM) yield and botanical composition were determined in 2014 and 2015. Results showed that plantain-containing mixtures significantly increased DM yield by on average 9.5% (20% plantain in seed mixture) to 13.6% (60% plantain) compared to the ryegrass-clover mixture, while other mixtures with forb species produced yields similar to that of the ryegrass-clover mixture. These effects were independent of slurry application and consistent over two years. Moreover, plantain-containing mixtures produced higher yield than chicory- and caraway-containing mixtures, through greater biomass of plantain and/or complementary effects on red clover. These findings firstly demonstrate that increasing species diversity through including certain competitive forbs promotes production of ryegrass-clover mixtures.

Keywords: plant diversity, forb, *Plantago lanceolata*, competition, dry matter yield, fertilization

Introduction

Plant productivity is often found to increase with increasing plant species/functional groups in natural ecosystems (Cardinale *et al.*, 2012).Thus, there is growing interest in exploring the role of crop diversity in enhancing agricultural productivity and sustainability (Tilman *et al.*, 2002). In European temporary grasslands, there are usually only a few crop species, which are dominantly by perennial ryegrass, red clover and white clover (*Trifolium repens* L.), since they can complement N use and produce high yield (Rasmussen *et al.*, 2012). Recent studies show that three forb species (chicory, caraway and plantain) are very competitive when grown with ryegrass-clover mixtures, providing the potential of further improving the productivity of the latter (Søegaard *et al.*, 2011). Therefore, this study aimed to experimentally examine the potential of these three forbs, individually and together, when grown with ryegrass-clover mixtures and whether the potential depends upon slurry application level. We hypothesize that inclusion of these competitive forbs will improve or maintain the production of ryegrass-clover leys, depending on the competitiveness of the forb species.

Materials and methods

A field experiment with three replicates was established in spring 2013. In each replicate, two levels of cattle slurry (0, 250 kg total N ha^{-1} year^{-1}) and ten seed mixtures were arranged in a two-way factorial design. The seed mixtures consisted of the perennial ryegrass-red clover mixture either alone or grown with different proportions of one or all of the three forb species (Table 1). Seed rates of each species in a mixture were calculated by multiplying their seed rates in pure stand (15, 4 and 12 kg ha^{-1} for perennial ryegrass, red clover and the three forbs, respectively) with proportions of the species in the mixture. Herbage biomass in the whole plot (1.5×8 m) was harvested four times (late May, early July, mid-August and early October) in 2014 and 2015 to determine annual dry matter (DM) yield. Botanical composition

Table 1. Plant species and their proportions in ten seed mixtures.

Plant species	GC	20CI	20CA	20PL	60CI	60CA	60PL	20CCP	60CCP	80CCP
	% of plant species in seed mixtures									
Ryegrass (GR)	50	40	40	40	20	20	20	40	20	10
Red clover (RC)	50	40	40	40	20	20	20	40	20	10
Chicory (CI)		20			60			6.7	20	26.7
Caraway (CA)			20			60		6.7	20	26.7
Plantain (PL)				20			60	6.7	20	26.7

of the mixtures was determined by hand separation of sub-samples into the five sown species and weeds. Annual DM yield over four harvests was analysed statistically using a linear mixed-effects model with replicate and plot as random effects, and with seed mixture, slurry application and experimental year as fixed effects. Differences between factor levels were tested using Tukey's *post hoc* test. All analyses were performed using the R software version 3.2.2.

Results and discussion

Annual herbage DM yield varied significantly ($P<0.001$) in the ten seed mixtures, showing a consistent pattern across two years (Mixture × Year: $P=0.13$) and across two slurry levels (Mixture × Slurry: $P=0.15$) (Figure 1). Compared to the ryegrass-red clover mixture (GC), inclusion of 20 or 60% plantain (20PL or 60PL) in seed mixtures increased DM yield by on average 9.5 or 13.6%, respectively, while other mixtures with one forb (chicory or caraway) or with all three forbs, produced similar DM yield. These results indicate that the role of crop diversity in promoting production of ryegrass-red clover mixture depends on forb species. Plantain-containing mixtures enhanced DM yield mainly because of larger biomass of plantain than caraway in both years (Figure 1) and chicory in 2015 (Figure 1C and 1D). Although chicory had comparably large biomass as plantain in 2014 (Figure 1A and 1B), the yield of red clover in chicory-containing mixtures was significantly lower than that *in planta*in-containing mixtures, suggesting that niche complementarity may occur between plantain and red clover.

The positive effect of plantain on red clover may be attributed to rosette leaves of plantain that stay close to the ground, allowing more light to be intercepted by red clover, whereas chicory may have strong competition for light on red clover through its large leaf area (Søegaard *et al.*, 2013). Red clover can in turn facilitate the growth of plantain by providing biologically fixed nitrogen (N) to plantain through rhizodeposition or mycorrhizal networks (Pirhofer-Walzl *et al.*, 2012). Indeed, N was a limiting factor in this study, as evidenced by enhanced annual DM yield (+9%) under slurry application (Figure 1). Slurry application significantly increased DM yield of ryegrass ($P<0.001$), did not or slightly increase yield of all the three forbs, but significantly decreased yield of red clover ($P<0.001$).

Recent studies have shown that forbs (chicory, caraway and plantain) produced higher mineral concentrations (e.g. Zinc) than grasses and legumes (Pirhofer-Walzl *et al.,* 2011), suggesting that including forbs in ryegrass-clover mixtures can not only enhance herbage production but also improve animal nutrition by providing sufficient dietary mineral supply to ruminants.

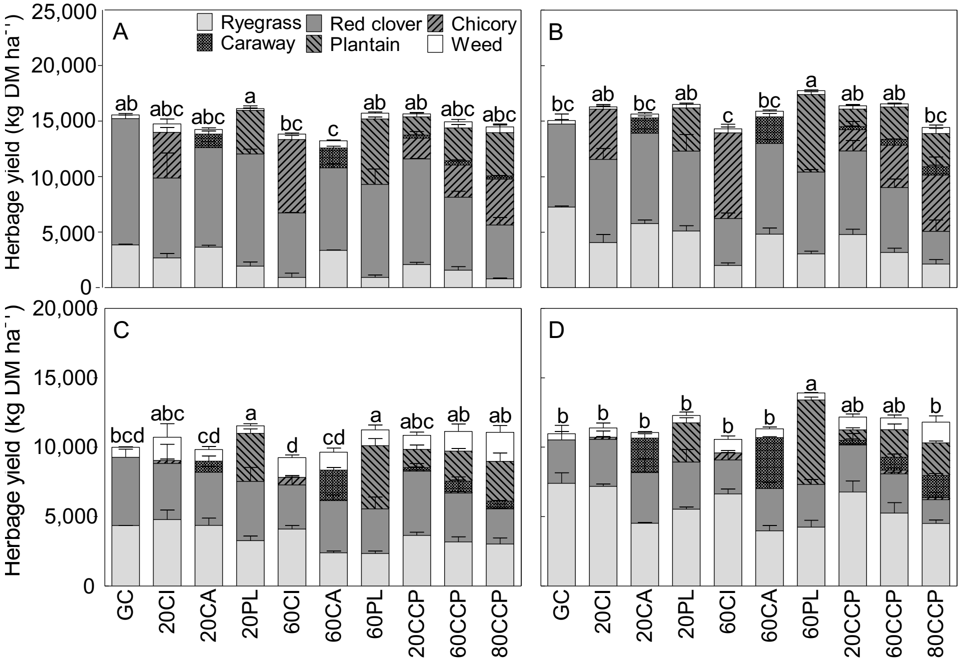

Figure 1. Annual dry matter (DM) yield in ten seed mixtures under two slurry application levels across two years. (A) Without slurry in 2014; (B) with slurry in 2014; (C) without slurry in 2015; (D) with slurry in 2015. Data are means ± standard deviation for each component species (n=3). Abbreviations of ten mixtures in the X-axis refer to Table 1. Means with different lowercase letters are significantly different (P<0.05) using Tukey's *post hoc* test.

Conclusions

We firstly demonstrate that including an additional functional group (i.e. forb) in traditional high-producing ryegrass-red clover mixtures improves or maintains productivity, depending on the competitiveness of specific forb species. Plantain-containing mixtures improve productivity, which can be a promising strategy for enhancing agricultural productivity and forage quality in European temporary grasslands.

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Successional change after grassland abandonment

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Abstract

The present agricultural landscape reflects a long history of changing land-use and farming practices, caused by e.g. technological development, urbanization processes and climate changes. A deeper understanding of how agricultural practices have altered the landscape is essential for the management of biodiversity and conservation of semi-natural grasslands. In this study, we explore the influence that changes in agricultural land-use, and grassland abandonment, have on successional changes in vegetation. The distribution, patch size, and plant species composition of semi-natural grasslands in a Central Norwegian agricultural landscape were mapped during two summers. Semi-natural grassland species decreased from managed grasslands to late regrowth successional phase, while number of forest species increased. Structural changes, e.g. increasing litter and tree cover, were also seen along the succession. Variation in species composition was related to management intensity and successional phase along the main gradient.

Keywords: semi-natural grassland, species richness, land use change

Introduction

Preindustrial agricultural land-use in Norway was characterized by mowing and/or grazing without ploughing and normally also without use of fertilizer. The semi-natural grasslands provided fodder for the livestock, both in summer (outfield pastures) and in winter (hay-making outfields). From the beginning of the 20th century, the use of semi-natural grasslands has gradually diminished. In this project, we explore the influence changes in agricultural land use during the last 60 years have on semi-natural grassland vegetation, focusing on effect of grassland abandonment on species richness and vegetation structure.

Materials and methods

Semi-natural grassland in 76 patches were mapped in a Central Norwegian upland area (ca. 7 km^2). A grassland patch was defined as a patch of min. 100 m^2 with semi-natural vegetation of same type according to the typification system 'Nature types in Norway' version 2.0 (Halvorsen *et al.,* 2015). The surveyed grasslands constituted a chronosequence ('space for time') that we used to examine the successional process from managed to late successional grasslands. Sampled grassland patches included 23 patches with present managed (grazed or mowed), 11 patches in fallow, 32 in early regrowth successional phase and 10 in late regrowth successional phase. Patch size ranged from 142 m^2 to 3,143 m^2. Successional phases were estimated based on the presence of young trees or shrubs indicating an ongoing succession, combined with information on land cover change seen on aerial photos from 2014 and 1960s.

All vascular plants were registered in each patch, and the frequency of each plant species was estimated on a scale from one to six. In addition, the canopy cover and cover of shrubs, plants in the field layer, bare soil, mosses/lichens and litter in each patch were registered as their percentage cover. The vegetation was also characterized using variables representing the impact of grazing (five classes; no grazing to intensively grazed), soil fertility (five classes; little to highly fertile soil) and a variable representing the long term management intensity from habitats with little influence of agricultural management to intensively managed grasslands (three classes).

Species richness was measured as the total number of species per patch. The results for species richness are presented as estimated least square means from linear models with successional phase and grassland area as explanatory variables. Pielou's evenness index, J, was calculated as J = H/log(S), where H is Shannon's diversity index and S is number of species. Species are characterized as typical for semi-natural grasslands and forests based on the background documents for 'Nature types in Norway' (NiN) version 1.0. Detrended correspondence analysis (DCA) was used to explore the variation in the species composition and how this variation is related to measured environmental variables. All analyses were performed using R and the package *vegan* (Oksanen *et al.,* 2015).

Results and discussion

The total number of species registered in grassland patches was 176 (including 6 to genus). The number of species per patch ranged from 33 to 71. The numbers of species were similar in all successional phases when adjusted for differences in patch size (Figure. 1). Grassland species decreased during succession, while number of forest species increased. The species composition in late regrowth phase is to a large extent a result of a mixture of species from open grasslands and forests. Evenness decreased from managed grasslands to early regrowth phase, and increased in late regrowth phase but the differences were only minor. Ceased management also entails changes in vegetation structure. In the late regrowth phase, the canopy became more closed, the field layer more thinned out and the cover of litter decreased. Also the moss cover decreased from managed and fallow phase to early and late regrowth phases. Bare soil had low cover in all successional phases. However, there were large variations in the cover of bare soil among managed grasslands, as these also included some patches with high grazing pressure and trampling.

The biplot from DCA ordination indicates that successional phase and environmental variables related to successional change, e.g. field layer and tree cover, are of key importance to explain the variation in the dataset (Figure 2). These variables are correlated with the 1st ordination axis (eigenvalue 0.193). The 2nd ordination axis (eigenvalue 0.142) is harder to interpret but the biplot indicates that this axis is related to the effect of grazing. Species characteristic to semi-natural grasslands tend to be in the centre and the left end of the 1st ordination axis representing successional change. This is in accordance with results from other studies (e.g. Gustavsson *et al.,* 2007).

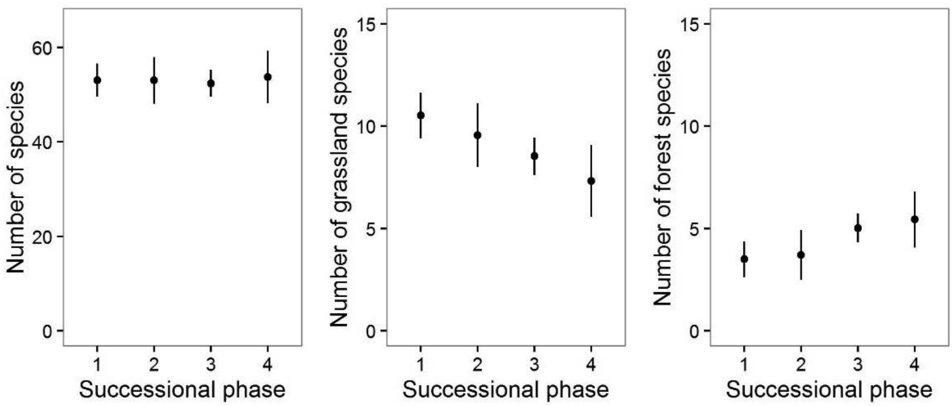

Figure 1. Least square means for number of vascular plant species (left), semi-natural grassland species (middle) and forest species (right) per patch in different successional phases (1: managed, 2: fallow, 3: early regrowth successional phase, 4: late regrowth successional phase). Vertical lines indicate 95% confidence interval. Note different scaling of y-axis.

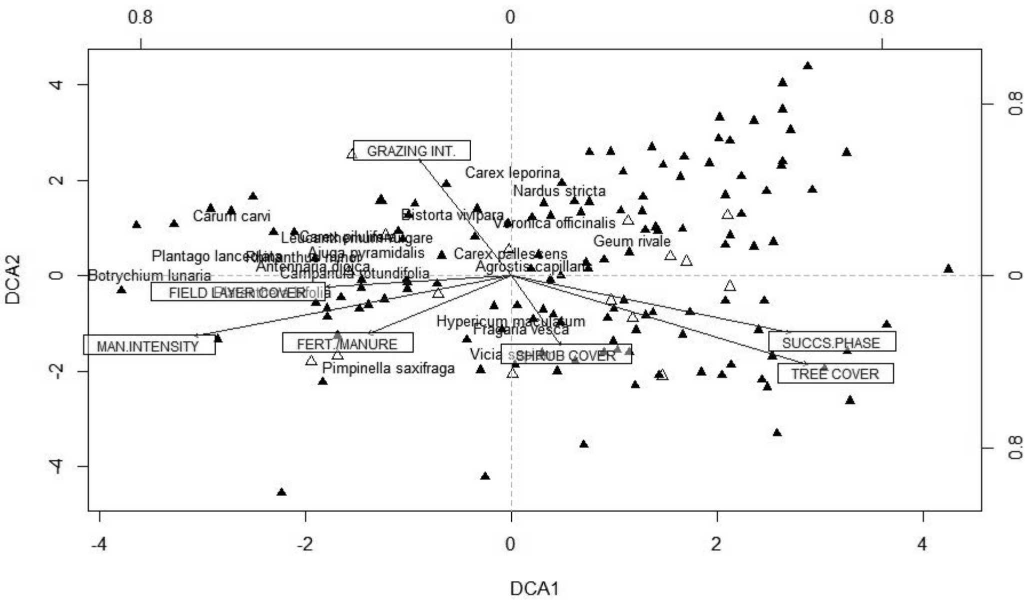

Fig. 2. Detrended correspondence analysis ordination of 76 semi-natural grassland patches. Biplot of species and environmental variables. Semi-natural grassland species are written with scientific name. Open triangles indicate forest species and filled triangles all other species. Abbreviations for environmental variables: man.intensity = management intensity, fert./manure = influence of fertiliser/manure, grazing. int = grazing pressure, succs.phase = successional phase.

These results show that the structure and species composition of semi-natural grasslands changes when agricultural management changes, although there are no or only minor changes in the overall number of species with the temporal and spatial scales examined in this study.

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Effect of long-term intensive and extensive grazing on plant species composition

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Abstract

To study effects of grazing intensity on temperate *Agrostis capillaris* grassland a long-term grazing experiment was established in 1998. The following treatments have been applied: (1) extensive grazing (EG), stocking rate adjusted to achieve a mean target sward surface height of more than 10 cm; (2) cutting in June and extensive grazing subsequently (ECG); (3) intensive grazing (IG), stocking rate adjusted to achieve a mean target sward surface height of less than 5 cm; (4) cutting in June followed by intensive grazing (ICG); and (5) unmanaged grassland (U). Since then, plant species composition has been recorded in fixed botanical squares each spring. Tall forbs (*Aegopodium podagraria, Galium album, Senecio* aggr., *Urtica dioica*) as well as tall grasses (*Alopecurus pratensis, Elytrigia repens*) had higher abundance in the U treatment and their cover reflected the management intensity U>EG, ECG>IG, ICG. Prostrate herbs (*Bellis perennis, Cerastium holosteoides Ranunculus acris*, *Veronica serpyllifolia*, *Trifolium repens)* were supported by both intensively managed treatments (IG, ICG). Both cutting treatments had significantly higher number of plant species than only grazed treatments (ICG>IG>ECG>EG>U). For the restoration of grassland biodiversity, hay cutting with aftermath grazing is the most effective treatment.

Keywords: grassland, heifers, grazing intensity, cutting, diversity

Introduction

Grasslands are an important part of the cultural landscape in Europe and their management has been affected by socio-economic conditions. Nowadays applied management should meet not only feeding requirements of animals but also provide other components of ecosystem services. Therefore long-term management experiments can help us to find a sustainable management acceptable for both farmer's profitability and nature conservation targets. Besides abiotic conditions, management is the key driver for the changes in plant species diversity. The impact of different grazing managements on vegetation dynamics has been widely studied in Europe during the last two decades (Pontes *et al.*, 2015). However, only a few studies were conducted more than five years, in which we can observe high variability in data caused by the resilience of presented plant species to various managements (Pavlů *et al.*, 2007). In this study we analysed intensive and extensive grazing effects on plant species diversity during 18 years of imposed treatments.

Materials and methods

The experiment has been undertaken since 1998 in the Jizerské hory mountains in the northern Czech Republic, 10 km north of the township of Liberec (50°50' N, 15°06' E). The site is underlain by granite bedrock and medium deep brown soil (cambisol) with the following attributes: pH_{KCl} = 5.1, C_{ox} = 3.9%, available P content = 64 mg kg^{-1}, available K content = 95 mg kg^{-1} and available Mg content = 92 mg kg^{-1}. The altitude is 420 m a.s.l., the mean annual precipitation is 803 mm and the mean annual temperature is 7.2 °C (Liberec meteorological station). The dominant species at the start of the experiment were *Agrostis capillaris*, *Alopecurus pratensis*, *Festuca rubra* agg., *Aegopodium podagraria* and *Galium album.* The long-term grazing experiment called the 'Oldřichov Grazing Experiment' (OGE) was established in the spring of 1998 and arranged in two randomized blocks (Pavlů *et al.*, 2007). Each block consisted of five paddocks where the following treatments were applied: (1) extensive grazing (EG), stocking rate adjusted to achieve a mean target sward surface height of more than 10 cm; (2) cutting in June and extensive grazing subsequently (ECG); (3) intensive grazing (IG), stocking rate adjusted to achieve a mean target sward surface height of less than 5 cm; (4) cutting in June followed by intensive grazing (ICG); and (5) unmanaged grassland (U). Each grazed paddock was circa 0.35 ha and the U paddock was 0.12 ha. All grazed paddocks were continuously stocked by young heifers (initial live weights of 150-250 kg) in each grazing season from early May until late October. Mean productivity of the pasture varied from 2 to 4 Mg DM ha^{-1} year^{-1}. The sward surface heights were measured weekly across each experimental plot (100 measurements) and stocking density was adjusted accordingly, by increasing or decreasing the area available for grazing. Permanent 1×1 m plots were analysed in four replications in each paddock. We recorded the proportional cover of all vascular species in early May each year from 1998 to 2014. Redundancy analysis (RDA) in the CANOCO 5.0 program (terBraak and Šmilauer, 2012) was used to evaluate multivariate vegetation data in the year 2014. The blocks were used as covariables to restrict permutations into blocks. A mixed linear model with block as random factor was used for evaluation of: (1) the number of vascular plant species in the year 2014 (one way ANOVA); (2) the cover of *A. capillaris* and *Trifolium repens* in the course of the experiment (repeated measures ANOVA).

Results and discussion

After 16 years of the experiment a diversification of plant species composition was revealed based on treatment application. The results of RDA from the year 2014 showed significant (P<0.001) effects of treatments for the first ordination axis and all ordination axes in plant species composition. The percentage of explained variability by the first axis and all ordination axes was 48.3 and 56.4, respectively. Tall forbs (*A. podagraria, G. album, Senecio* agg.*, Urtica dioica*) as well as tall grasses (*Elytrigia repens, Holcus mollis*) had higher abundance in the U treatment (Figure 1) similarly as ten years ago (Pavlů *et al.,* 2007), showing the stability of plant species composition in unmanaged grassland. Species associated with all managed plots regardless of the treatments (IG, ICG, EG, ECG) were *F. rubra* agg.*, A. capillaris, Poa trivialis, Ranunculus acris, Rumex acetosa, Plantago lanceolata* and *Taraxacum* spp. The cover of short grass *A. capillaris* and prostrate legume *T. repens* reflected defoliation intensity IG, ICG > EG, ECG >

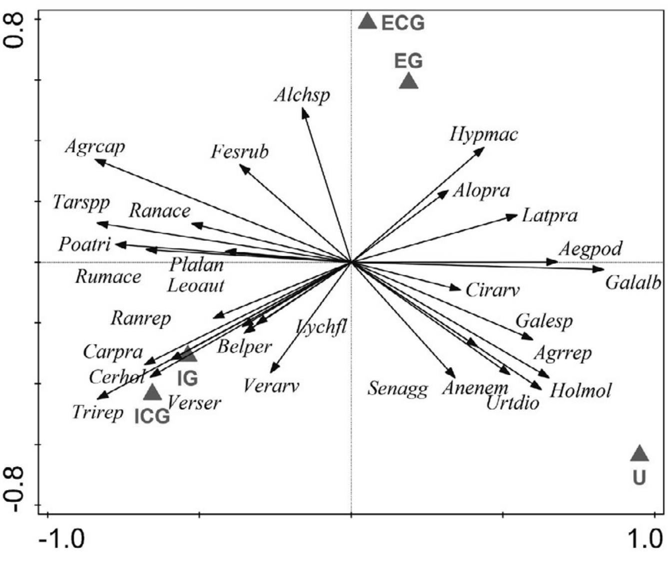

Figure 1. Ordination diagram showing the result of redundancy analysis analysis of vegetation data. For treatment abbreviations see Materials and methods section. Species abbreviation is based on the first three letters from genus and from species name.

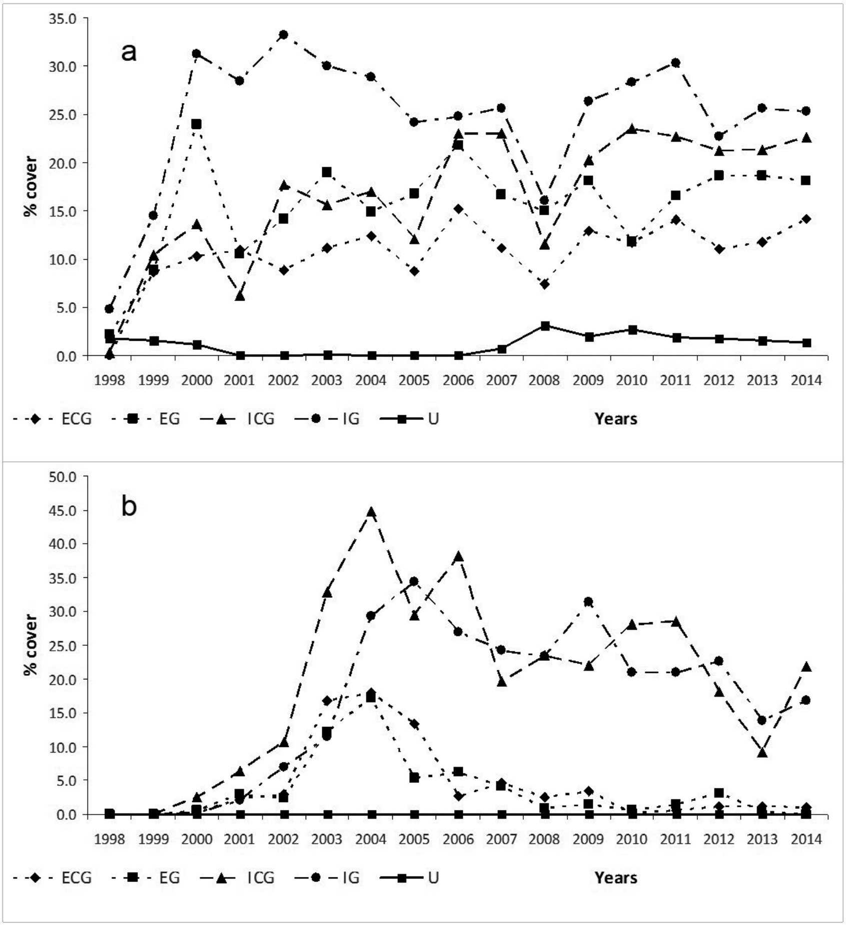

Figure 2. Changes in cover (%) of (A) *Agrostis capillaris*, (B) *Trifolium repens* for different treatments in the years 1998-2014.

U and was affected by treatment, time and their interaction (P<0.05) (Figure 2A, B). Prostrate herbs (*Bellis perennis, Cerastium holosteoides*, *Ranunculus acris*, *Veronica serpyllifolia*, *T. repens*) were supported by both intensively managed treatments (IG, ICG) (Figure 1). The mean number of vascular plant species in the year 2014 was 24.1, 23.8, 21.2, 19.8 and 17.2 for ICG, ECG, IG, EG and U, respectively. Although botanical compositon of IG and ICG as well as EG and ECG was relatively similar in dominats, both cutting treatments gave a significantly (P<0.001) higher total number of plant species than grazed-only treatments. Our results confirmed previous studies that management intensity is the key driver in terms of diversity of vascular plant species (Hejcman *et al.*, 2010; Pavlů *et al.*, 2011) and long-term grazing experiments are indispensable for further testing.

Conclusions

Further diversification of plant species composition based on treatment application was revealed after 10 years of the first evaluation of the Odřichov grazing experiment. However, it seems that some degree of equilibrium under each treatment was achieved, as the dominat species had a relative stable cover in the last five years. For the supporting of grassland biodiversity, hay cutting with aftermath grazing was the most effective treatment.

Acknowledgements

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How can livestock farmers contribute to maintaining and increasing ecological networks?

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Abstract

Rural landscapes containing dense hedgerow networks and permanent grassland have diminished in France, as a result of agricultural intensification and homogenization. These relatively extensively managed habitats are considered important for biodiversity conservation and current policy aims to conserve and restore such habitats through implementation of ecological networks. To achieve this aim, it is important to understand how increasing hedgerow density and permanent grassland area will challenge livestock farmers, and also to understand how groups of farmers at landscape scale can interact to enhance ecological continuity. In the Vendée 'bocage', in western France, we questioned farmers individually about their willingness to enter into a number of management scenarios and then we conducted a group interview to assess possibilities for shared action. While the capacity to increase hedgerow length is above all influenced by sociological factors, increasing the proportion of permanent grassland needs to be thought out in relation to farm strategy; different levels of farmer adaptability were identified. The co-construction of coherent hedgerow networks seems to be a more realistic goal than the establishment of permanent grassland continuity. These results are discussed in the light of a recent fieldwork study, highlighting the ecological value of these habitats, for nesting birds.

Keywords: hedgerow, permanent grassland, biodiversity, livestock, farm strategies, collective action.

Introduction

Rural landscapes containing hedgerow networks and permanent grasslands have diminished in France, as a result of agricultural intensification and homogenization (Huyghe *et al.*, 2014). As these areas provide habitats for wildlife, current legislation aims to conserve, restore and connect them via the implementation of ecological networks. But managing, planting or destroying hedgerows and/or grassland depend above all on individual farmer decisions. Like Gibon (2005), we assumed that such decisions would mainly relate to the expected function of these habitats from a strategic farming point of view. For example, we know that the ways in which farmers view grasslands can vary considerably: while little or no value may be attached to unwanted or poor-quality grasslands, wanted and managed grasslands can be perceived more positively, providing opportunities for workforce optimisation, lower inputs, higher incomes and better animal performance. Such perceptions are closely related to soil and climate conditions and to farm production strategies (Peyraud *et al.*, 2009). We supposed that individual farmers would vary in their ability to envisage increases in the area of land allocated to hedgerows or grasslands at farm-level. However, even if certain farmers are ready to make changes, the successful implementation of ecological network policy will depend on collective action at a regional scale involving numerous individual farms. Local authorities responsible for conservation policy implementation are currently inciting farmers to integrate ecological planning considerations into their farming strategy. Our aims were to better understand how potentially increasing hedgerow density and areas of permanent grassland area would challenge livestock farmers' strategy and to explore how groups of farmers at landscape scale might interact to enhance ecological continuity.

Materials and methods

We selected a rural borough located in Vendée (western France) dominated by cattle farming but with a low density of hedgerows and grasslands in comparison with the surrounding region. Twenty cattle farmers (among 40 in the borough) were interviewed about (1) farm production strategy, (2) social perceptions of hedgerows and grasslands, (3) farmer's social position and (4) ecological network scenarios. Regarding the fourth point, our aim was to determine how willingly farmers would adapt to increasing hedgerows and grasslands and so we suggested two prospective scenarios: (1) re-designing the plot arrangement to create plots up to 2.25 ha in area entirely surrounded by hedges, (2) re-organizing the crop system to obtain at least 50% of the farmed area as permanent grassland. For each scenario, the respondent had to propose changes on a map representing his farm and indicate the prerequisites for their success (none, changes in farming practices, with or without farm strategy modification). Based on the answers, farms were grouped according to farmer adaptability. We interpreted the relationships between these differing levels of adaptability and farm strategies, hedgerow/grassland perceptions and farmers' social position. Following this, all forty livestock farmers within the borough were invited to a collective workshop. Twelve farmers participated, of which 11 had been previously surveyed. Then farmers were split into 3 groups, each group of farms representing a continuous area of farmland. On a local map, and using pawns of different colours, farmers were asked to: (1) locate hedgerows and permanent grasslands; (2) identify the hedgerows and grasslands networks discontinuities; (3) propose collective scenarios to reinforce hedgerows and grasslands continuities. The maps thus generated were photographed and conversations recorded. Debate contents were analysed to highlight the factors enabling ecological network construction and their effects on farming practices/strategies.

Results and discussion

Individually, none of the farmers were willing to fully implement the suggested hedgerow scenario. Three did not want to plant any more hedges while the others (n=17) were willing to plant hedgerows, upon certain conditions, within and around permanent grassland plots. These attitudes were mainly related to sociological factors and were poorly explained by farm strategy. The willingness to plant hedgerows was stronger when farmer perception of hedgerows was positive and multifunctional and indeed when younger farmers were involved in the decision-making. On the other hand, on farms which had undergone major restructuring including hedgerow removal, and where perception of hedgerow function was negative (loss of productive land, maintenance costs, etc.), motivation for new planting was naturally limited. When questioned on the same theme in a group, several changes in attitude were observed, though the suggested objectives were less challenging in comparison to the individual survey. In particular: (1) farmers were able to reach a consensus on hedgerow discontinuity locations and restoration; (2) farmers who had claimed they would be very willing to plant hedgerows restrained their ambitions, proposing simple restoration along existing boundaries, roads and tracks; (3) new conditions needed for hedgerow restoration to be possible were cited, with regard to the relationships between farmers and local authorities, neighbours and landowners. The agricultural functions of hedgerows and farming strategy were never mentioned during these debates.

As far as permanent grassland was concerned, three of the surveyed farmers had already achieved the 50% grassland scenario. Six farmers felt unable to increase the areas of permanent grasslands on their farms. Among the others, 4 would be ready to make small changes to crop rotation practices without any change to farm strategy, while 7 would be able to change farm strategy (de-intensification of animal production, decreased crop production in favour of grasslands). Grasslands were considered by all respondents to be an important tool for reaching production objectives and rarely in terms of environmental benefit. Willingness to consider permanent grassland restoration depended mainly on the agricultural function of these areas, closely related to long-term farm strategy. For instance, farmers who had come through an economic crisis who were aiming to reduce inputs cited increasing grassland area as the main method for reaching this goal. Conversely, farmers who had developed intensive animal production (saturated agricultural area, buildings, debt capacity and workforce) had no intention of increasing permanent grassland areas. In the collective workshop, farmers were not able to imagine coordinated strategies for permanent grassland restoration, as modifying the amount of permanent grassland strongly affects individual farm strategy. In some groups, however, farmers did share information about the roles of grassland in their farming system.

To conclude, the workshop showed as far as hedgerows concerned, that it is possible to constitute groups of farmers able to debate the local improvement of such networks. However, the equivalent was not true for grasslands, due to their perceived importance for production objectives. In order to overcome this difficulty, it may be possible to organise local debates on the role of grasslands within farm strategies, and thereby identify ways to reinforce grassland networks at local and regional scales (Opdam *et al.*, 2015). If farmers find it potentially easier to envisage increases in hedgerow density as opposed to grassland area, this may be good news for certain bird communities. Our bird surveys in Vendée have shown that breeding bird communities are concentrated in hedgerows, which support a relatively high species richness and abundance of generalist or forest specialist species. The possible drawback is that specialist farmland birds of greater conservation concern prefer more open habitats.

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Tree-livestock interaction promotes nutrient shift and influences plant species richness in orchards

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Abstract

Orchards are traditional but endangered agroforestry landscape elements in the continental part of Europe and of high importance for nature conservation. Trees introduce diversity into grasslands, as they establish microsites with different conditions through modification of light availability and soil chemical parameters. In orchards, livestock may promote distinct nutrient deposition as they tend to rest and defecate under trees, which might affect plant species composition and richness. In 42 orchards within the Rhenish Uplands (Germany) we studied how different grazer species (cattle, horses and sheep) affect soil nutrient accumulation (P_2O_5 and K_2O) in microsites along a tree-proximity-gradient (under the crown, ecotone, non-tree affected area) and how this influences plant species richness in microsite habitats and at paddock scale. Our results show higher amounts of K_2O and P_2O_5 and fewer numbers of species under the crown compared to the ecotone and non-tree affected area. Further, we found differences in plant species richness at the paddock scale among different grazer species with higher diversity on horse compared to cattle and sheep grazed pastures. Increasing grazing intensity promoted K_2O deposition and decreased species richness at microsite and paddock scale.

Keywords: nutrient deposition, grazer species

Introduction

In the Continental European landscapes, orchards are traditional but endangered agroforestry elements with high nature conservation value. Usually, they experienced no history of agricultural improvement and they therefore display relatively high plant species richness. Furthermore, trees introduce diversity into grasslands, as they increase the richness of niches with considerable variation in light availability and soil chemical parameters, enabling coexistence of species with different requirements at the microscale (Garbarino and Bergmeier, 2014). Despite grazing animals differ in their grazing impact, they are known to enhance grasslands dynamics, diversity and species richness at all scales by selectivity, trampling and nutrient deposition (Ollf and Ritchie, 1998, Rigueiro-Rodríguez *et al.,* 2009). Therefore, grazers may enhance tree-dependent spatial heterogeneity due to nutrient deposition as they tend to rest and defecate under trees in orchards (Wilson *et al.,* 2007). Soil nutrient concentration in general has been shown to affect plant species richness and composition (Janssens, 1998) but there is a lack of research on the combined effect of grazing induced nutrient shift and plant species richness in orchards. Therefore, in this study we test the hypotheses that in orchards grazer species differ in their impact on (1) heterogeneity in soil nutrient concentration of microsites, (2) species richness of microsites and (3) species richness at paddock scale, and that these effects also depend upon grazing intensity.

Materials and methods

In 2014 we studied a total of 42 grazed orchards within the Rhenish uplands in Germany. Sites were arranged in a nested triplet design, comparing horses, cattle and sheep grazing at similar site conditions (altitude, soil and slope). Within each site 3 trees were chosen randomly and at each tree 3 microsites (1 m^2) were arranged under the crown ('trunk'), in the ecotone and in an area that was not tree-affected ('open'). Soil samples (500 ml) were collected at a depth of 10 cm, pooled for microsite categories respectively and analysed for plant available phosphorus (P_2O_5) and potassium (K_2O). All plant species per microsite were identified to determine species richness (SR) at microsite. SR at paddock scale was defined as cumulative number of species found in the nine microsites. Data on site-management were recorded interviewing farmers using standardized questionnaires. As a variable for grazing intensity, live weight-unit (500 kg) grazing days per year were calculated for each site and standardized per hectare. Statistics were performed in R 3.2.2 (R Development Core Team 2008). Linear Mixed Effects Models (lme) were applied to analyse the effects of microsites, grazer species and grazing intensity on soil pH, P_2O_5, K_2O and species richness at micro scale and paddock scale using nlme package. SR at paddock scale was tested for effects of grazer species and grazing intensity. In all models tree nested within site and, this, nested within triplet was considered as a random effects structure. Minimal adequate models were assessed performing stepwise backward selection with χ^2-test and significant parameters were estimated by Maximum Likelihood Estimation. Assumptions were tested graphically.

Results and discussion

We found significant differences in soil chemical parameters between microsites indicating tree and grazing induced nutrient deposition within paddock. Concentrations of P_2O_5 and K_2O showed lower values with increasing distance to the tree (Figures 1A, B) which is consistent with findings of Wilson *et al.* (2007). Hypothesis 1 is confirmed, as a significant interaction of microsite with grazer species was obtained.

Close to the trunk the concentrations of P_2O_5 were significantly higher in cattle than in horse or sheep grazed paddocks ($P<0.001$) and K_2O concentrations were significantly higher in cattle and sheep compared to horse paddocks ($P<0.001$). Further, the concentration of K_2O is significantly ($P=0.049$) modified by grazing intensity. Generally, horses are known to establish distinct latrine areas where they urinate and defecate and introduce distinct heterogeneity in grasslands swards and nutrient deposition (Schmitz and Isselstein, 2013; Wrage *et al.,* 2011) whereas cattle and sheep tend to drop their excreta more homogenously (Ollf and Ritchie, 1989). In orchards, nutrient deposition seems to differ in this

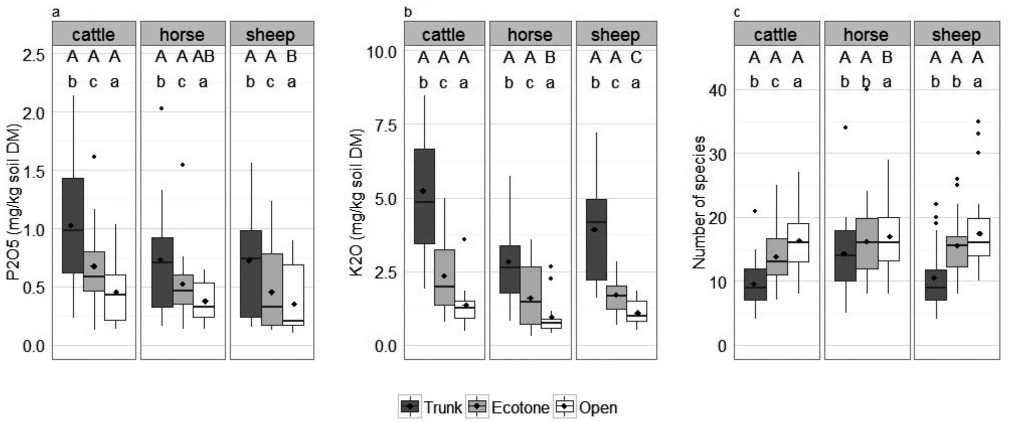

Figure 1. Allocation of nutrients (A) P_2O_5, (B) K_2O and (C) species richness at the microsites 'Trunk', 'Ecotone' and 'Open' of cattle, horse and sheep paddocks. Interactions of microsite and grazer species were significant for P_2O_5 ($P<0.001$), K_2O ($P<0.001$) and SR ($P<0.001$). Significant differences among grazer species within microsites are represented by upper case letters and of microsites within grazer species by lower case letters. Dots within the bars represent mean values. DM = dry matter.

respect. Horse latrine areas are not necessarily depending on the presence of trees while cattle use the shady tree areas for resting and obviously also for defecation and urination.

In general, the number of plant species increased significantly with increasing distance to the tree and decreasing nutrient concentration (Figure 1C). Yet, this effect was modified by the grazer species as SR of the trunk area was significantly highest on the horse paddocks while there was no difference among the grazer species in the ecotone and open microsite. This is in accordance with hypothesis 2. Janssens *et al.* (1998) showed distinct negative effects of P_2O_5 concentrations on species richness which is in line with our data of P_2O_5 concentration and number of species at the microsites. On paddock scale SR was significantly affected by grazer species (P<0.001), with slightly higher SR in horse grazed pastures, but also significantly modified by grazing intensity (P<0.001).

Conclusions

In orchards plant species richness is affected by nutrient deposition, grazer species and grazing intensity. Horse grazed pastures showed slightly higher species richness than cattle or sheep grazed orchards. Therefore, concerning conservation of plant species richness in orchards, in absence of sheep or cattle, horses should also be reconsidered as grazer species for nature conservation strategies.

Acknowledgements

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Environmentally and economically sustainable dairy and beef production in Sweden

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Abstract

Dairy and beef systems are linked to each other. Therefore the potential for environmental improvement of these systems has to be analysed jointly. In this study, future environmentally sustainable supply chains for milk and beef were created using best practice and results from the best quartile of current production systems. These comprised cropping, livestock, processing industry, logistics, package and wastage, but not retailers or consumers, in a typical Swedish county. The current situation was described and three solution scenarios, focusing on ecosystem services, plant nutrients and climate impact, were created. Life cycle assessments and production cost calculations were performed for all four scenarios. The results revealed great potential to reduce negative and strengthen positive environmental impacts of Swedish dairy and beef production with maintained production volumes, irrespective of environmental aspect studied. Good agricultural practices, especially concerning feeds, were essential, but processing and distribution also contributed. All solution scenarios resulted in lower costs of production than today.

Keywords: sustainable food chains, LCA, food system scenarios, cattle, milk, beef

Introduction

In order to design more ecologically sustainable food production, it is necessary to study the entire supply chain and consider the economic sustainability of primary production. In an interdisciplinary project, Sonesson *et al.* (2015) attempted to create future environmentally and economically sustainable food chains embracing several production chains. The parts dealing with dairy and beef production are presented in this paper.

Materials and methods

Various experts collaborated in an interactive research process described by Sonesson *et al.* (2015). Supply chains for milk and beef in the Swedish county of Västra Götaland were studied, focusing on drinking milk/hard cheese and sirloin steak, respectively. Production systems reflecting the most common current production systems (in 2012) were designed and named 'Reference' scenario. Three environmentally improved systems were then developed using best practice and data from the best quartile of current production systems. These future solution scenarios aimed to deliver the same amount of commodities as today with improved environmental performance, while production economics were improved or kept at present levels. The scenarios were: 'Ecosystem', aiming at maintaining and developing ecosystems; 'Nutrients' aiming at optimised plant nutrient use and supply; and 'Climate', aiming at reduced climate impact. The environmental impacts of these supply chains were quantified and compared with the 'Reference' using life cycle assessment (LCA) for the separate product chains of drinking milk and sirloin steak and for the whole county cattle system. The costs in primary production of dairy and beef were calculated for the scenarios based on 2013 price levels and 2015 revised EU payments. The scenarios comprised:

- Ecosystem. High-yielding dairy production based on grass silage + complementary extensive beef production with heifers and steers on semi-natural pastures;
- Nutrients. High-yielding dairy production based on grass and maize silages + complementary indoor beef production based on maize silage;
- Climate. Moderate-yielding dairy production based on clover and maize silages and pasture + complementary indoor intensive beef production based on forage.

Results and discussion

According to the LCA calculations, good agricultural practices, especially for feeds, but also processing/distribution, contributed to reduced negative environmental impact in all solution scenarios compared to 'Reference'. The product chain for drinking milk generally performed better than 'Reference' (data not shown). However, for the product chain for sirloin steak (not shown) and the whole county cattle system (Table 1), the impacts in most environmental categories increased in 'Ecosystem', while 'Nutrients' also showed some increases. This was due to the higher proportion of calves originating from less environmentally efficient suckler cows as a result of higher milk yield per dairy cow and an assumed constant volume of beef. However, suckler cows use semi-natural pastures and in 'Ecosystem' their increased number managed more pastures, resulting in more biodiversity than in the other scenarios (Table 1).

For both dairy and beef, the most cost-efficient production models in the solution scenarios had considerably lower net costs than 'Reference' (Figures 1 and 2; 1 SEK = 0.11 EUR). The estimated total cost per kg milk was lowest in 'Ecosystem' and 'Nutrients' with their high-yielding cows. The costs were substantially higher in 'Climate', which assumed moderate-yielding cows. In beef production, the environmental payment for grazing semi-natural pastures was important for the net cost. In cases with high environmental payments, rearing models with steers and heifers in the 'Ecosystem' scenario with much grazing competed very well with the best indoor bull model in 'Nutrients' and 'Climate' (Figure 2). However, in cases with lower environmental payment for pasture, indoor bull models became more competitive (not shown). The dairy × beef models generally had lower costs than beef breed models.

Table 1. Total yearly potential environmental impact from dairy and beef production today ('Reference') and for three solution scenarios. Relative values where 100 = total emissions/acreage for 'Reference'.

	Global warming potential, ton CO_2-eq.	Acidification, kmol H^+-eq.	Freshwater eutrophication, kg P-eq.	Marine eutrophication, kg N-eq.	Cumulative energy demand, TJ-eq.	Acreage managed semi-natural pastures, ha
Reference dairy	58	64	64	52	72	19
Reference beef	42	36	36	48	28	81
Reference total	100	100	100	100	100	100
Ecosystem dairy	89	75	112	158	103	90
Ecosystem beef	115	105	123	145	148	222
Ecosystem total	100	86	116	152	116	196
Nutrients dairy	78	37	93	98	92	90
Nutrients beef	80	64	103	98	117	120
Nutrients total	79	47	96	98	99	115
Climate dairy	58	32	72	89	67	110
Climate beef	69	53	90	75	111	73
Climate total	63	39	78	82	79	81

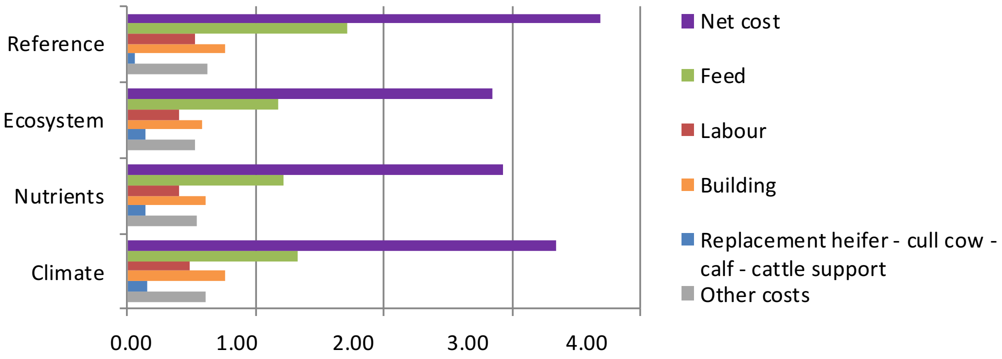

Figure 1. Cost of production (SEK kg^{-1} energy-corrected milk) for the four scenarios.

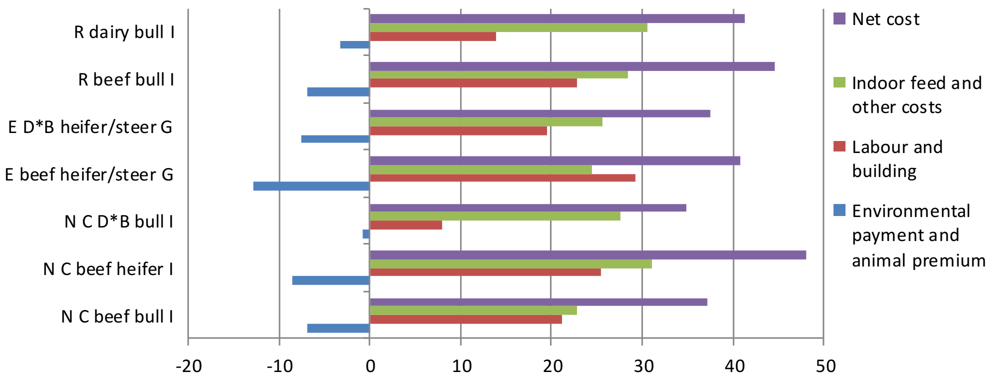

Figure 2. Cost of production (SEK kg^{-1} carcass weight) assuming all grazing occurs on semi-natural pastures where 1/3 of the acreage qualifies for high and 2/3 for basic payment. R is Reference scenario; E (Ecosystem), N (Nutrients) and C (Climate) are the three solution scenarios. D (Dairy) and B (Beef) refer to the breed of the calf; I is indoor rearing; and G is grazing during summer. For B, the costs of the dam are included.

Conclusions

There is a great potential to simultaneously reduce negative environmental impacts and decrease costs in Swedish dairy and beef production, whilst maintaining production volumes.

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Nitrogen fixation in red clover grown in multi-species mixtures with ryegrass, chicory, plantain and caraway

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Abstract

While many studies have investigated effects of species composition and management on N_2-fixation dynamics in simple clover-grass mixtures, there is a lack of knowledge about the performance of legumes and their N_2-fixation in more species-rich grassland including non-legume forbs. This study investigated N_2-fixation and total N yield in grassland mixtures including different combinations of red clover, perennial ryegrass and three deep rooted forbs: chicory, ribwort plantain and caraway, managed without and with slurry application. The percentage of clover N derived from atmosphere (%Ndfa) increased in mixtures, but was unaffected by inclusion of forbs. However, forbs affected the quantity of N_2-fixation and total N yield by affecting the red clover proportion in the mixtures. Mixtures composed of grass, clover and plantain or caraway showed higher N_2-fixation and N yield than with chicory included. Slurry application reduced N_2-fixation due to decreasing clover contents in the sampled biomass. The study highlighted differences among forbs in their influence on legume N_2-fixation and total N yield showing that forbs can be included in grasslands without negative effects on red clover performance.

Keywords: forb, grassland mixture, slurry, N_2-fixation, N yield

Introduction

Nitrogen is often the limiting nutrient for plant production, and the input of N from N_2 fixation in legume-rhizobia symbioses is widely considered as valuable sources of N in agroecosystem. Several studies have reported that plant species diversity of forage legume and non-legume in grasslands increases competition for available soil N, thereby stimulating legumes to increase their dependency on N_2-fixation and reducing the risks of losing N to the environment (Palmborg *et al.*, 2005; Rasmussen *et al.*, 2012). In addition, the introduction of non-legume forb in grasslands has been shown to increase the acquisition of nutrients from deep soil layers (Pirhofer-Walzl *et al.*, 2013) and improve forage quality (Pirhofer-Walzl *et al.*, 2011). The objective of this study was to determine how the inclusion of different forbs in different proportions in multi-species grasslands affects N_2-fixation in red clover and the total N yield of the mixtures.

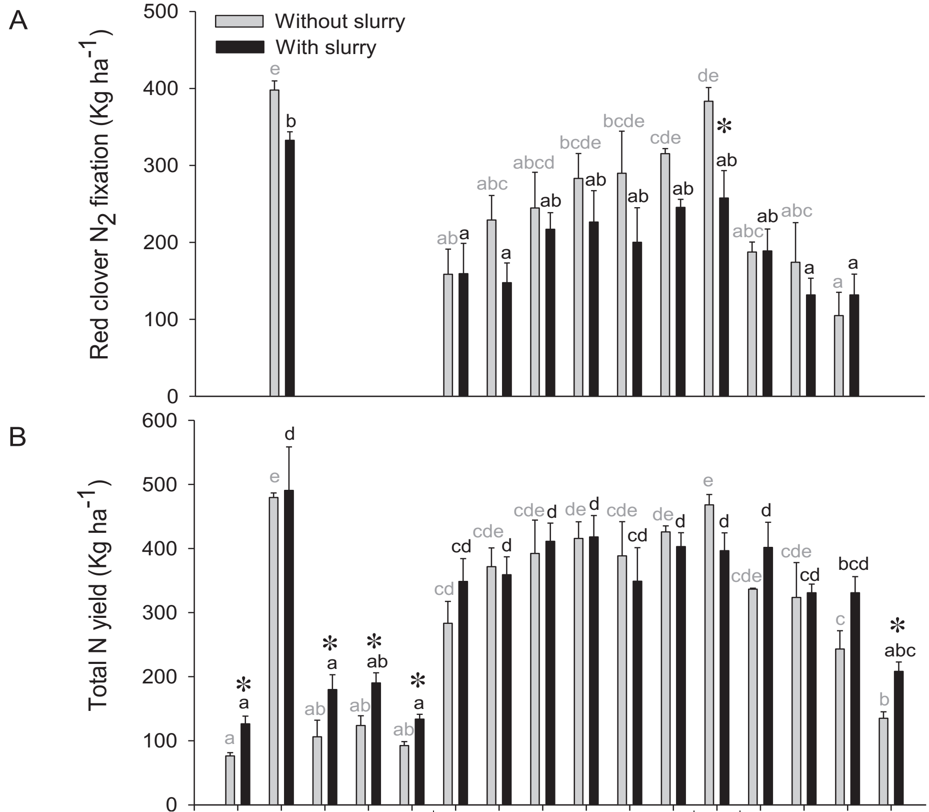
Materials and methods

The experiment was conducted at Foulumgaard experimental station, Aarhus University, Denmark in a sandy loam soil with a cropping history including both grassland and arable crops. Sixteen seed mixtures (Figure 1) composed of different combinations of perennial ryegrass (*Lolium perenne* L.), red clover (*Trifolium pratense* L.) and three forbs: chicory (*Cichorium intybus* L.), ribwort plantain (*Plantago lanceolata* L.) and caraway (*Carum carvi* L.) were sown in 1.5×8 m plots in spring 2013, in a replacement design based on proportions of each species seeding rate in pure stand, which were 15, 4 and 12 kg ha^{-1} for ryegrass, red clover and forbs, respectively. Each mixture was exposed to 0 vs 250 kg N ha^{-1} in the form of cattle slurry applied in 2014 in three replicates. N_2-fixation was quantified using the 15N isotope dilution method (Rasmussen *et al.,* 2012) after labelling a 1×1 m subplot in each experimental plot with ammonium sulphate (0.1 g N m^{-2} and atom fraction 15N = 98%) in early spring 2014. The aboveground biomass was harvested to 5 cm stubble height in each subplot four times during the growing season

(May – October). The samples were sorted to individual species, dried, weighted and analysed for total N concentration and atom fraction 15N. The N_2-fixation was quantified based on excess atom fraction 15N (subtracting the background 15N measured in plant samples from unlabelled plots) in legumes and non-legumes, and total N yield was quantified based on N concentration and aboveground biomass yields of all species in each mixture in the subplot.

Results and discussion

The whole season biomass yield ranged from 4.5 to 15.8 Mg dry matter (DM) ha^{-1} without slurry, and from 7.4 to 16.7 Mg DM ha^{-1} with slurry application (data not shown). Mixtures of ryegrass, red clover and forbs produced almost as high biomass yields as did the pure stand of red clover and standard mixture of ryegrass and red clover. The proportion of red clover in the biomass samples ranged from 30-82% without and 30-61% with slurry application.



Plant species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
	Pure stand					One forb species						Std.	Three forb species			
Perennial ryegrass	100					20	40	20	40	20	40	50	40	20	10	
Red clover		100				20	40	20	40	20	40	50	40	20	10	
Chicory			100			60	20						7	20	27	33
Ribwort plantain				100				60	20				7	20	27	33
Caraway					100					60	20		7	20	27	33

Seed mixture

Figure 1. Whole-season N_2-fixation (A) and N yield (B) of above ground biomass without and with slurry application (mean ± standard error; n=3). '*' indicates a significant effect of slurry application, and different letters indicate significant differences within each slurry level (P<0.05). '*Std.*' refers to the standard mixture of perennial ryegrass and red clover.

Red clover derived a significantly (P<0.001) higher proportion of its N from N_2-fixation (%Ndfa) in mixtures than in pure stand. There was no significant difference in %Ndfa between mixtures, which was above 90% even in the treatments which received slurry application. On an annual basis, red clover fixed between 104 and 398 kg N ha^{-1} without and from 132 to 333 kg N ha^{-1} with slurry application. The amount of N_2 fixed was highest in the pure stand of red clover followed by the standard mixture of grass and clover (Figure 1A). Red clover in three species mixtures composed of grass, clover and plantain or caraway fixed nearly similar amounts of N_2 as did the red clover pure stand and the standard mixture. The slurry application decreased the amount of N_2 fixed in the majority of the mixtures, and this effect was most pronounced in the standard mixture (P<0.01). The total whole season N yield varied between 76 and 479 kg ha^{-1} without slurry and from 126 to 491 kg N ha^{-1} with slurry application, nearly following the pattern of biomass production. The majority of the mixtures obtained similar N yields as did the red clover pure stand and the standard mixture (Figure 1B). Slurry application increased total N yield in pure stands of non-legumes and mixture of three forb species (P<0.05).

The variations in the amounts of N_2-fixation and N yield in the mixtures were caused mainly by different proportions of red clover, as influenced by the competitiveness of the forbs for biomass production. Chicory and plantain showed better competitiveness than caraway in the mixtures. Without fertilisation, total N yield decreased with high proportions of chicory in the seeded mixture, which showed that the decrease in N_2-fixation was only partly counteracted by the high productivity of chicory. These findings suggested that choice of species and proportion of the forb component are important considerations when designing diversified leys for efficient N use.

Conclusions

Forbs can be included in grassland mixtures without affecting N_2-fixation and total N yield, provided that the mixture does not contain too high a proportion of chicory, as this forb is a strong competitor that might reduce clover yield and N_2-fixation. These findings show promising possibilities for the design and implement of multifunctional, multi-species grassland mixtures, i.e. ones that combine the valuable N inputs from legumes with efficient N uptake in grasses and deep-soil nutrient acquisition and high forage quality of forbs.

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Variation of legume contents and symbiotic nitrogen fixation under intensive grazing

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Abstract

Legumes are important components of pastoral systems because of their ability to fix N_2 and because of their high nutritional value. We investigated the effects of soil, land-use history, grazing intensity and weather conditions on legume content in temperate grassland in a 13-year-long study (including the centennial drought year 2003 and wet years) on six permanent ('old') and six sown ('new') pasture paddocks, covering a large range in soil properties. Species' contributions to aboveground biomass were assessed on permanent quadrats three times every year (1,355 relevés). Old and new grassland approached a near-equilibrium legume contents with half-lives <2 yr, irrespective of initial legume content. At equilibrium, legume content (on average: 4%) and N fixation (on average: 6 kg ha^{-1} yr^{-1}) were low. The centennial drought in 2003 caused a 'drought crash' of legumes followed by a fast recovery. In the long term, neither weather nor soil properties (water holding capacity, nutrient levels), or grazing intensity had a distinct influence on legume content under conditions of low N losses and continuous N recycling under permanent grazing and no additional nutrient inputs from fertilizers or concentrate feeding.

Keywords: *Trifolium repens*, clover, succession, drought, spatio-temporal variation

Introduction

Legumes are important for pastoral systems because of their ability to fix N_2 and because of their high nutritional value. However, unpredictability of legume content and low legume contents cause reluctance to rely on legumes as the predominant source of N particularly in view of the fact that the relation between legume contents and pasture productivity is still not fully understood. We examined the influence of management (sowing and grazing intensity), weather (including a centennial drought) and a wide range of soils (varying in water and nutrient availability) in a long-term experiment.

Materials and methods

We conducted a 13 year-long study (including the centennial drought year 2003 and wet years) on 12 pastures exhibiting a large range in plant available water (PAW) capacity. Half of the pastures were situated on old permanent grassland and half were sown at the beginning of the experiment. Sowing mixtures comprised *Trifolium repens* and *Trifolium pratense*, which contributed 9 and 2% of the number of seeds). All pastures were grazed continuously during the vegetation period without additional feeding. Grazing pressure was adjusted to three levels (for details on pasture properties and management see Schnyder *et al.,* 2006). On each pasture four permanent monitoring quadrats (1 m^2) were installed (marked by buried magnets). Each quadrat was subdivided into 100 tiles (10×10 cm^2). Species' percentage in aboveground biomass, plant height, soil cover and other parameters were visually recorded on each tile, which allowed quantifying species contribution down to 0.01% on a quadrat. Total biomass was estimated by two trained, regularly 'calibrated' persons. Measurements were taken three times every year (early, mid and late growing season), yielding a total of 1,355 relevés. Symbiotic N fixation was estimated by an empirical model (Høgh-Jensen *et al.,* 2004).

Results and discussion

Shortly after seeding, legume content on newly sown pastures was about three times higher than on old pastures (Figure 1, left panel) exceeding the percentage in seeds by a factor >4. On both types of pastures, legume content quickly decreased over time while the swards adapted to the new and constant grazing regimen. Decreases followed exponential functions with half-lives of legume contents of 0.7 and 1.3 yr. After about 10 years, both types of pastures approached the same and very low legume content (about 4% of above-ground biomass), which accounted for a rate of N fixation of on average 6 kg ha^{-1} yr^{-1}.

Legumes were dominated by the two sown species on the new grassland, but *T. pratense* quickly disappeared and *T. repens* remained. On the whole, *T. repens* was the only legume species on old grassland. During the last years, spontaneous species (*Lotus corniculatus, Medicago lupulina, Trifolium dubium*) established in the sown grassland but their overall contribution to total community biomass remained low (<0.5% when combined).

During the extreme drought year (2003, 4th year after establishment of the experiment), legume contents decreased dramatically but the lowest contents were found early in the growing season of the following year although rainfall was above average in 2004 (Figure 1, middle and right panel). The drought crash was most pronounced on the new pastures, which had a lower plant PAW capacity than old pasture (on average 70 mm vs 140 mm) and consequently a lower mean actual PAW during the growing season in 2003 (8 mm vs 15 mm).

In mid-summer 2004, legume content started to recover and reached its maximum early in the growing season 2006 on both pasture types. During late 2005 and early 2006 legume content even exceeded the values predicted from the long term trend, compensating the low values during 2003 and 2004. Thereafter, the legume contents returned to the long-term trend.

The variance of legume contents between quadrats strongly decreased over time as a consequence of approaching a very low equilibrium value (Figure 2, both left panels). This was true for the spatial variation (between quadrats) and temporal variation (between measuring events). Also it was true for old and new pastures although the decrease was less pronounced on old grassland, which had started at lower legume contents that also allowed for less initial variation. The inter-annual variation contributed most to total variance during the first years, when legume contents declined quickly; later inter-annual variance added only little to total variance, indicating that weather conditions – including the centennial

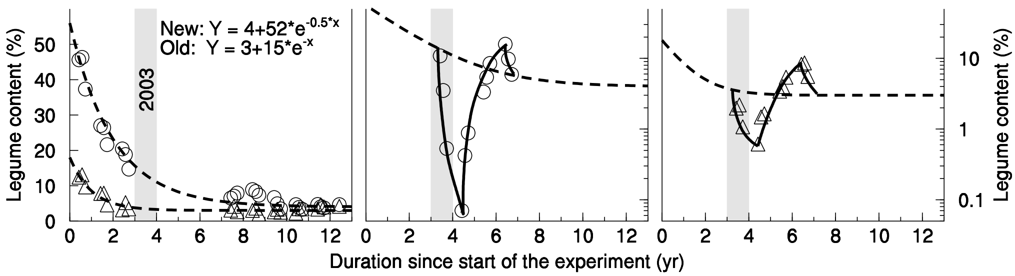

Figure 1. Mean legume contents averaged for sampling dates on new (○) and old pastures (Δ). The left panel displays long term trends without the drought crash, while the middle and right panels display contents during the centennial drought (2003, grey shaded) and the recovery years (2004-2006) on a logarithmic y axis. Dashed lines in panels give the long-term exponential trends. Markers are means of all quadrats within six pastures.

Figure 2. Variation in legume content over time and space. The experimental period was divided in four periods (T1 = 2000-2003, T2 = 2004-2006, T3 = 2007-2009 and T4 = 2010-2012). The left pair of panels displays the contribution of space (hatched) and time (grey) to total variance on new and old grassland while the right pair displays the relative contribution of inter-annual (dark grey), intra-annual (light grey), within-pasture (hatched) and between-pasture (cross hatched) variation to total variance.

drought in 2003 – had a minor impact on variation of legume contents (Figure 2, both right panels). The intra-annual (seasonal) variation was very low (<10%) at all times on both new and old pastures. While the relative contribution of temporal variation declined strongly over time, the relative contribution of spatial variation increased strongly (>90% during the last term). The within-pasture variation showed no clear pattern over time, while the variation between pastures contributed most to the total variation during the last phase of the experiment. Still, total variance and thus also variance between pastures was extremely small during the last phase (SD 2%) indicating that differences in grazing pressure (compressed target height during grazing: 4 to 7 cm; mean cattle mass: 475 to 830 kg^{-1} ha^{-1} yr^{-1}), in PAW capacity (varying between 59 and 186 mm), soil nutrient supply (ranging from low to very high for P and K according to the German recommendations) and soil N pool (ranging from 4,400 to 9,700 kg ha^{-1} in the top 10 cm of the soil) had only a minor influence on legume content.

Conclusions

Under experimental conditions mimicking real-farm management, legume contents adapted quickly to permanent grazing irrespective of the initial values (half-lives <2 yr). On the long-term, neither weather (even including a centennial drought) nor soil (PAW capacity, nutrient levels), or grazing intensity distinctly influenced legume content. In all cases, legume contents and N fixation rates became low under conditions with little losses and continuous N cycling by permanent grazing even without additional N input from fertilizers or concentrate feeding.

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Upscaling soil carbon and nutrient losses from dairy farms to regional level

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Abstract

An important aspect of agro-ecology is adjusting farm management to the regional landscape. In intensive farming systems, the approach may target, among others, reduction of N- and P-losses along water borders and/or an increase biodiversity through cultivation of cereals. However, not much is known of the short- and long term impact of such measures on soil carbon contents (SOC). An ex-ante assessment was made of soil carbon change following implementation of measures to reduce nutrient losses in the region of Winterswijk (NL). The Roth-C model was used to assess carbon loss over time. Upscaling to farm and regional level was done on the base of potential area sizes. It was found that, although individual measures that reduce N- and P-losses may lead to either loss or gain in SOC-content at field level, implementation of all measures would at the regional level lead to zero change in SOC-stock. Thus in theory it is possible to improve water quality while maintaining SOC-contents at the regional level. However, this result depended largely on the area sizes where some high impact measures may be implemented, e.g. cultivation of cereals and raising pH. It was concluded that discussing farm and regional priorities for soil and water quality could be the next step in the regional stakeholder process.

Keywords: N and P losses, Roth-C model, dairy farms, regional level

Introduction

An important aspect of agro-ecology is adjusting farm management to the regional landscape. This may involve agricultural practices to realise production goals, e.g. drainage. It may also involve practices directed at several external targets, e.g. reducing N- and P-losses to improve water quality and/or increase biodiversity. Governing bodies focus primarily on the accumulated effect of measures for achieving the regional targets. Effects of these measures on soil carbon contents (SOC) in agricultural soils are seldom taken into account. A regional stakeholder process that includes both farm and landscape qualities is important if intensive dairy farms are to maintain nature values (Hanegraaf *et al.,* 2015). For the present study, we performed an ex-ante assessment of soil carbon change related to successful measures for reducing nutrient losses and increasing biodiversity. Results at field level were scaled up to the regional level to provide data and insights as possible input for the regional stakeholder process.

Materials and methods

The work was carried out in the Winterswijk region in the Netherlands. This region is characterised by a 'coulissen' landscape on sandy soils, consisting of a mosaic of agricultural lands, hedgerows and woodlots, dissected by brooks and rivers. Dairy farming is the dominant farming system. In previous studies, farmers have been testing measures to reduce N- and P- losses and improve biodiversity (Den Boer and De Haas, 2013). The applicability of successful measures throughout the region was assessed using technical data (groundwater level, pH) and farmers' evaluation. It was found that for the 5,000 ha area, nutrient losses could be reduced by 123 t N and 72 t P_2O_5, which equals 9 and 20% of the amounts used, respectively. Successful measures were taken as a starting point for modelling changes in SOC-contents at regional level. It concerned the following: (1) cultivation of grain cereals instead of maize or potato; (2) introduction of grass-clover mixtures; (3) cultivation of a good catch crop after maize; (4) no farmyard manure (FYM) if soil P is high (c. 7 P-PAE) within 10 m of water borders; and (5) liming to raise pH to recommended level in acid maize soils. The change in SOC-content with each measure was assessed using the Roth-C model (Coleman and Jenkinson, 1999) and a time-scale of 20 years. The outcomes of the modelling at the field level were used for upscaling to the regional level, using the data of area sizes from the previous study.

Results and discussion

It was shown that the selected measures to reduce N- and P-losses may lead to either loss or gains in soil carbon content (Figure 1). As a starting point, a SOC-content of 65 Mg ha^{-1} was assumed, which evolved widely over a period of twenty years, i.e. from 49 Mg C ha^{-1} (potato without FYM to 74 Mg C ha^{-1} (cereals, incorporating straw).

Results showed a gain in C for both grassland and grass-clover; however without FYM, SOC-contents remain stable. The assessment was based on calculations with a standardised C-input of a 3-year old grassland which reflects actual agricultural practice in the region. As the new CAP stimulates permanent grasslands, gains in C may be even higher. The measure 'liming' illustrates that not all measures that reduce N- and P-losses turned out positive for SOC-contents. Over a period of 20 years, C-loss would be c. 8 Mg ha^{-1}, corresponding to a change on soil organic matter content from 3.3 to 2.8%. A possible consequence of this measure could be that SOC contents decrease to the extent that other related soil properties may become limiting, e.g. soil moisture. During the research period, farmers experienced a positive effect of this measure on crop yield, which was concomitant with higher N- and P-uptake. It could be argued from an agronomic viewpoint that this measure should be applied at all relevant fields, in combination with application of extra organic matter. Other measures were found to increase SOC-contents, e.g. the cultivation of cereals to provide concentrates at dairy farms. It was calculated that this measure would increase soil organic matter content from 3.3 to 3.8% over the 20-year period. This measure performed well for three aspects, i.e. reduction of nutrient losses, increase of biodiversity, and increase of SOC. However, chances that this measure will be implemented throughout the region are small, since it has not been included in the new CAP. Dairy farmers with min. 75% grassland and max. 30 ha arable land are exempt from the diversification requirement. We therefore expect that dairy farmers with derogation (80% grassland) will resume to grass and maize cultivation.

At the regional level, it was found that maximum implementation of all measures concerned would lead to almost zero change in SOC (Hanegraaf *et al.,* 2016). Thus for the region of Winterswijk it was found that water quality may be improved without deteriorating soil quality. This result was largely dependent

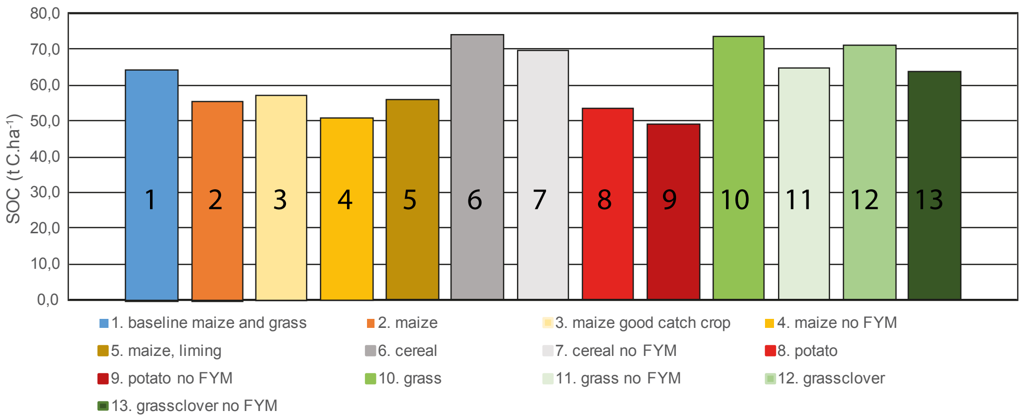

Figure 1. Changes in soil carbon-content (SOC) after 20 years.

on the area sizes where high impact measures could be implemented, e.g. cultivation of cereals and raising pH. Other selections of measures and area sizes may lead to different results concerning losses or gains in SOC-content at regional scale level. Discussing targets at the regional level may help in selecting priorities for adjusting farm management with respect to a reduction of nutrient losses while maintaining soil fertility.

Conclusions

This work focused on improved adjustment of farm management on critical levels of the regional landscape, e.g. soil type, groundwater level, and hedges. The approach showed that, at the regional level, it is possible to reduce N and P losses while maintaining SOC levels. Several combinations of measures are feasible, depending on farm characteristics as required roughage production, economics, and effect of individual measures on SOC. Next steps in the regional stakeholder process could be setting farm and regional priorities and agreement on measures to implement with or without a rewarding scheme.

Acknowledgements

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Phytoextraction of soil phosphorus by grass-clover as a synergy between agriculture and nature restoration

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Abstract

In the development of the Dutch National Ecological Network, arable land is converted to nature areas. This development is often hampered by high phosphorus (P) levels. Standard practices to decrease the amount of P are either top-soil removal or mowing of low yielding established grassland. Both methods have their disadvantages and there is a need for additional techniques. As an alternative, phytoextraction ('mining') of soil P has been proposed. We tested a phytoextraction by cropping an intensively mown grass-clover with potassium (K) fertilisation, which could potentially be used as cattle feed. A long-term field experiment was conducted, comparing soil P removal by grass-clover swards with, and without, K fertilization on a sandy soil. Our results show that grass-clover with K fertilization is an effective method that removed excess soil P at a relatively high rate (34 kg P ha^{-1} year^{-1}, significantly higher than without K fertilization, P<0.05). Therefore phytoextraction can be an important synergy between agricultural production and nature restoration.

Keywords: grass-clover, potassium fertilisation, phytoextraction phosphorous, nature

Introduction

In the Netherlands, an extensive network of green corridors, called National Ecological Network is being implemented. In this context, many hectares of arable land have been transferred to nature reservations. In most of these former agricultural soils, long-term applications of fertilisers and animal manure in the past, that have stopped after land acquisition by nature organizations, have led to an imbalance in the levels of soil nutrients: while these soils often have high levels of relatively immobile soil P, the more mobile nitrogen (N) and potassium (K) have often become suboptimal according to agricultural standards. Apart from problems with leaching and eutrophication of surface water, high soil P levels are a threat to certain types of species-rich plant communities. One of the methods to decrease soil P levels for nature restoration is topsoil removal, but this practice is often expensive and conflicts with objectives of soil conservation and conservation of historical heritage. Hence, there is an urgent need for a better strategy to remove excess P from former agricultural land in a way that ensures successful development of 'ecosystem target types', such as nutrient-poor, species-rich grasslands. We designed a method to test phytoextraction of P with grass-clover swards. Experiences in practice in various nature areas using grass-clover swards resulted in 'losing' the clover from the sward and very low dry matter (DM) yields after some years. We argued that legumes are weak competitors for K and hypothesized that (1) K limitation could be the cause of clover decline in grass-clover swards in nature areas, and that (2) with K fertilisation, grass-clover swards could be an effective method to reduce soil P levels to levels low enough to accommodate species-rich plant communities. We designed a field experiment with a duration of 7 years to test these hypotheses, with the aim to develop a new and cost-effective way to remove excess P from former agricultural soils, by phytoextraction of P with a grass-clover sward (Timmermans and Van Eekeren, 2016).

Materials and methods

In a field experiment, two treatments were compared: (1) no K fertiliser application, and (2) K fertiliser application at a rate of 398 kg K ha^{-1} per year. Our experiment was set up on a former arable field that is currently part of a nature conservation area. The field with the experiment had a noncalcareous sandy soil (Spodosol; Podzol, cover sands of pleistocene origin). Some soil characteristics were: 5.8% soil organic matter in the topsoil, pH-KCl of 4.3 and K-HCl 28.2 mg K kg^{-1}. The experimental plots were sown in 2002, and treatments and measurements continued until 2009. The experimental design was a randomized block design in 4 blocks, with 4 replicate plots (4×10 m) per treatment. All plots were sown with a mixture of 30 kg ha^{-1} perennial ryegrass (*Lolium perenne*) and 4 kg ha^{-1} white clover (*Trifolium repens*). At each harvest date, all plots were mown with a two wheel tractor (Eurosystems P55). Freshly mown material was weighed and a representative fresh sample of about 700 g was dried at 70 °C for 24 hours. A sub-sample of these dried samples was analysed for P and another sub-sample was dried at 105 °C for one hour additionally to determine DM content and calculate total DM yield. Soil samples from the 0-10 cm soil layer were taken each year in winter and analysed for total P and ammonium lactate extractable P (P-AL). Statistical analysis was performed using GenStat. Repeated measures ANOVA was used for comparing annual data across years and treatments.

Results

In the first two years of the experiment the total DM yield (clover plus grass) was similar between treatments, but significantly lower in the unfertilised plots from 2004 onwards, up until the end of the experiment in 2009 (Figure 1).

The amount of P removed by grass-clover was significantly higher in the K fertilised treatment than in the unfertilised treatment (34 kg P ha^{-1} $year^{-1}$ compared to 26 kg P ha^{-1} $year^{-1}$ on average over the years). Compared to differences in DM yield, differences in P removal were somewhat smaller between treatments, because P content (g kg^{-1} DM) in the treatment without K fertilisation increased with decreasing biomass (P<0.001; R^2=0.18). Chemical analyses of soil samples from the experimental plots showed significant decreases in P-total and P-AL in both treatments over the years (Figure 2). Soil P levels were generally lower in K fertilised plots than in unfertilised plots, but this treatment effect was not significant (P=0.085 and P=0.107).

Discussion and conclusions

Our results indicate that our first hypothesis was correct: in plots without K fertilization, productivity of grass-clover was lower than in K fertilised plots. Our results also support the first part of our second hypothesis, i.e. that grass-clover combined with K fertilisation can be an effective method to reduce soil P levels in sandy soils: this treatment removed 34 kg P ha^{-1} $year^{-1}$ on average. Our results also provide

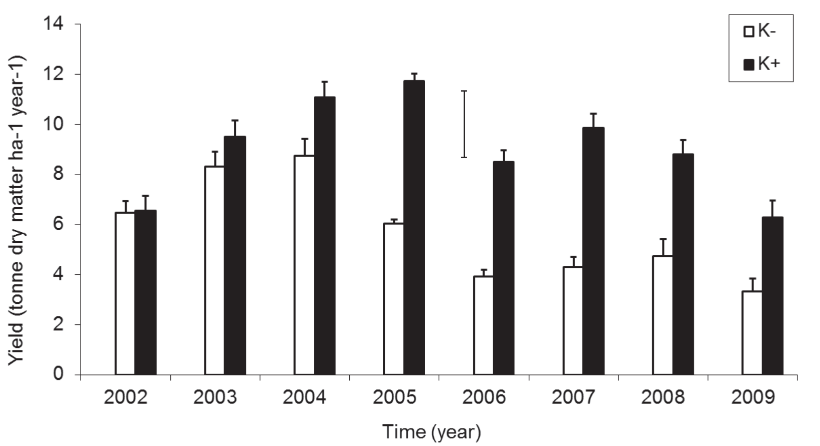
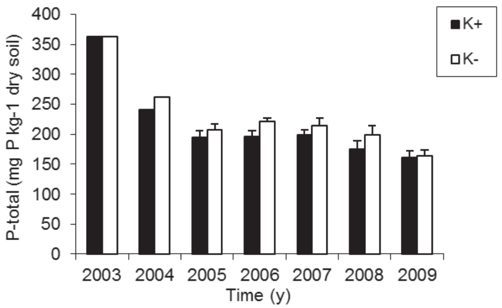


Figure 1. Average annual dry matter yield (t ha^{-1}) (P=0.004) in K fertilised plots and unfertilised plots, from 2002 to 2009. Error bars indicate standard error of the mean. The error bar at the top indicates the least significant difference (P<0.05) between the treatments.



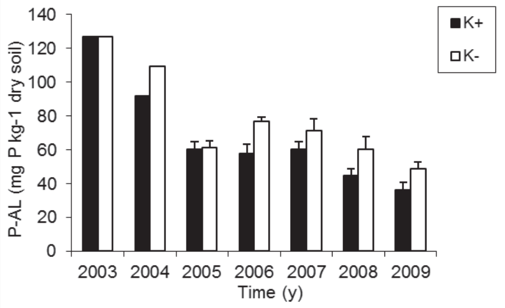


Figure 2. Average P-total (left) and P-AL (right) in the 0-10 cm soil layer of K fertilised plots and unfertilised plots.

evidence for the second part of this hypothesis: that this method could indeed remove enough P to arrive at a plant available P concentration low enough for species-rich grasslands. By the end of the experiment the average soil P level in the K fertilised treatment was comparable to soil P levels in nutrient-poor, species-rich grasslands in the region. We conclude that intensively harvested grass-clover fertilised with K is an effective alternative to topsoil removal to reduce excess P. Moreover, this method provides an elegant way to recycle P, i.e. to transfer excess soil P from new nature areas back into the agricultural system by cattle feed (Timmermans and Van Eekeren, 2016).

Acknowledgements

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Patch-dependent herbage growth drives paddock productivity in a long-term extensive grazing system

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Abstract

In extensive grazing systems selective grazing creates stable mosaic structures consisting of frequently grazed, short patches and rarely grazed, tall patches. The stocking rate determines the share of different patch types on a pasture. In the present experiment biomass production of different patch types ('short', 'medium', 'tall') and biomass production of paddocks treated with different stocking rates ('moderate', 'lenient', 'very lenient' – 6, 12, 18 cm target sward height) were analysed. The experiment was conducted on a long-term low-intensity set-stocked cattle pasture. We focused on total annual aboveground net primary productivity (ANPP) and on the temporal distribution of ANPP. We hypothesized that short patches are more productive and biomass growth is distributed more homogeneously throughout the season than in tall patches. Furthermore we assumed that paddocks grazed moderately are more productive and biomass growth is distributed more homogeneously than in more leniently grazed paddocks. The results showed that tall patches were not less productive than short patches, which led to similar ANPP of paddocks. ANPP of short patches and moderately grazed paddocks was distributed most homogeneously throughout the season.

Keywords: cattle pasture, ANPP, patch grazing, sward productivity, spatial heterogeneity

Introduction

In low-intensity, continuously stocked grazing systems selective grazing creates a stable mosaic structure out of frequently defoliated (short) patches and rarely defoliated (tall) patches (Dumont *et al.*, 2012). Kept in an early phenological stage (Richards *et al.,* 1962), short patches are known to be more productive than tall patches (Şahin Demirbağ *et al.*, 2008). Because stocking rate determines the proportion of different sward patch types on the pasture area (Tonn *et al.*, 2014), it is assumed that productivity of pastures grazed with different stocking rates differs from each other. On a long-term low-intensity experimental cattle grazed pasture with paddocks varying in target sward height, the effect of the stocking rate on aboveground net primary productivity (ANPP) of different sward patch types and paddock was studied. On the patch level, we hypothesized ANPP of short patches to exceed that of tall patches and biomass growth to be distributed more homogeneously. We further assumed that ANPP of paddock increases with increased stocking rate and that biomass growth is distributed more homogeneously

Materials and methods

The experiment was conducted on a low-intensity permanent cattle pasture located in Lower Saxony, Germany (51°46'N, 9°42'E, 180-230 m a.s.l.). Since 2005 grazing has been performed with three different stocking rates, defined by target compressed sward heights (CSH; Castle, 1976) in paddock: 'moderate grazing' (MG, 600 kg ha^{-1} a^{-1}), 'lenient grazing' (LG, 340 kg ha^{-1} a^{-1}) and 'very lenient grazing' (VLG, 220 kg ha^{-1} a^{-1}), with 6, 12 and 18 cm CSH, respectively, measured via rising-plate-meter. The experiment is arranged in a randomized block design and made up of three replications (9 paddocks à 1 ha each). Target sward heights are kept constant via bi-weekly sward height measurements (50 per paddock) and adding or removing cattle in response (put-and-take system). Since 2013, three different sward patch types have been defined according to seasonally adapted CSH class thresholds: 'short' (SP), 'medium' (MP) and 'tall' (TP). One exclusion cage (2×1 m^2) per patch type and paddock was (re-) placed throughout the vegetation period, resulting in 6 (7) growth periods in 2013 (2014) with length adapted to seasonal biomass productivity. CSH was measured at two 0.25 m^2 quadrats per cage, pre and post cage (re-)placing, respectively. Standing herbage mass was predicted from a given CSH by using a calibration model previously set up for the experiment. Herbage growth from each pre to post CSH measurement date and subsequently mean daily growth rates for each combination of patch type and paddock within each growth period were calculated. For the calculation of cumulative herbage growth (ANPP), negative daily growth rates were set to zero. $t_{0.5ANPP}$ is defined as the date at which half of the total annual ANPP was produced, representing the distribution of herbage growth. The share of different patch types on each paddock area was determined via bi-weekly CSH measurements, hence ANPP for each paddock could be calculated. All statistical analyses were carried out with R 3.2.3 (R Core Team, 2015). Data were fit with linear mixed effects models using the package 'nlme'. Treatment means, their standard errors and the significance of their pairwise differences (Tukey test) were estimated using the package 'lsmeans'.

Results and discussion

Results are shown in Table 1. On the patch level there was a significant interaction between patch type and stocking rate (P=0.005) on ANPP. $t_{0.5ANPP}$ differed significantly between patch types (P<0.001). For all three stocking rates SP showed a significantly lower ANPP than TP. The results are in contrast to those of Şahin Demirbağ *et al.* (2008), who found a higher productivity for SP on the same experimental pasture in the beginning of the experiment. This may be based on soil nutrient depletion in SP due to long-term nutrient uptake by grazers within inter-annually stable vegetation patterns, especially on pastures with low grazing pressure (Dumont *et al.*, 2012). TP accumulated a great amount of biomass in early spring, reaching $t_{0.5ANPP}$ significantly earlier (P<0.001) than MP and SP, for which herbage growth was distributed more continuously throughout the season. The result is in line with our hypothesis. In TP herbage senescence rates become higher than growth rates with increasing sward age (Binnie and Chestnutt, 1994) leading to a higher share of dead and reproductive plant material (Şahin Demirbağ *et al.*, 2008) than herbage of frequently grazed SP. Better rainfall distribution in summer and autumn 2014 postponed $t_{0.5ANPP}$ for all patch types in MG to a later date (P=0.036). The same was true for paddocks, for which $t_{0.5ANPP}$ under MG was reached latest in 2014, differing significantly (P=0.0402) from all three stocking rates in 2013. A more homogeneous nutrient uptake and higher proportion of the paddock area covered by excreta under intensive grazing (Moir *et al.*, 2011) is likely to lead to a more homogeneous distribution of soil nutrients within the pasture area in MG. Hence, the good weather conditions in 2014 may have mobilized soil nutrients, making them available for plant growth in autumn.

Table 1. Annual aboveground net primary productivity (ANPP) in t dry matter per ha and the day from April 1st at which half of ANPP is reached ($t_{0.5\ ANPP}$) for three different sward patch types and paddocks under different stocking rates. Shown are estimated means and standard errors of three replications.

Year	Stocking rate	Patch type 'short'		Patch type 'medium'		Patch type 'tall'		Paddock	
		$t_{0.5\ ANPP}$	ANPP	$t_{0.5\ ANPP}$	ANPP	$t_{0.5\ ANPP}$	ANPP	$t_{0.5\ ANPP}$	ANPP
2013	Moderate	48 (9)	5.8 (0.7)	44 (4)	10.5 (1.1)	41 (2)	9.4 (1.1)	46 (3)	7.5 (0.2)
	Lenient	56 (5)	3.8 (0.7)	47 (2)	6.1 (0.8)	38 (1)	9.6 (1.0)	42 (3)	7.4 (0.9)
	Very lenient	63 (8)	3.3 (0.7)	48 (3)	4.7 (0.7)	39 (1)	7.4 (0.9)	40 (3)	7.3 (1.6)
2014	Moderate	88 (6)	6.5 (0.7)	85 (12)	10.8 (1.0)	69 (10)	8.3 (0.8)	84 (8)	8.5 (0.7)
	Lenient	70 (3)	4.5 (0.7)	61 (9)	6.4 (0.7)	41 (6)	8.4 (0.9)	51 (8)	7.0 (0.8)
	Very lenient	82 (5)	4.0 (0.6)	68 (11)	5.0 (0.7)	47 (9)	6.2 (0.7)	47 (8)	5.5 (1.1)

Conclusions

Because ANPP of SP was lower than that of TP, a soil nutrient depletion in SP may be assumed. On paddock scale similar ANPP under different stocking rates represent ANPP of patch types influenced by long-term application of grazing treatments. Good rainfall distribution in 2014 altered $t_{0.5ANPP}$ in MG on patch and paddock scale, indicating a mobilization of more homogeneously distributed soil nutrients under this grazing treatment.

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High functional dispersion of forage mixtures suppresses weeds in intensively managed temperate grassland

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Abstract

Weeds in managed grassland can result in substantial losses of forage yield and quality. We studied whether weeds could sustainably be suppressed through functionally diverse grassland mixtures with high resource use and biomass yield. Seedlings of five common weeds were established as phytometer plants in forage leys. Ley stands comprised of monocultures and mixtures with two and four species of *Lolium perenne* L., *Cichorium intybus* L., *Trifolium repens* L., and *Trifolium pratense* L., from which five plant functional traits (PFTs) were determined: leaf dry matter content, specific leaf area, leaf nitrogen (N) and carbon (C) concentration, and the C:N ratio. Phytometer plants were increasingly suppressed with increasing functional dispersion (calculated from PFTs) and biomass yield of ley stands, so that in the functionally most diverse ley mixtures the survival of phytometer plants was 48%, while in the functionally least diverse stands survival was 65%. In parallel, biomass yield was significantly greater in the functionally most diverse mixtures than in the average of monocultures with zero functional dispersion. Taken together, the results suggest that phytometer suppression occurred through increased capture of resources in functionally diverse ley mixtures. We conclude that growing forage mixtures instead of monocultures enhances competition against weeds and can be a sustainable strategy to improve weed suppression.

Keywords: *Cichorium intybus*, forage mixtures, *Lolium perenne*, plant functional traits, *Trifolium pratense*, *Trifolium repens*, weed suppression

Introduction

Weeds in managed grassland can result in substantial losses of forage yield and quality (Naylor, 2002). Although weeds can be controlled by herbicides, such measures come along with economic and environmental costs. In mixed grasslands for example, application of herbicides can affect sward composition, e.g. loss of dicotyledons including legumes (Suter and Lüscher, 2011), which results in limitations in biomass and nitrogen yields (Nyfeler *et al.*, 2009; Suter *et al.*, 2015). Recent attempts to achieve more sustainable grassland management include the use of multi-species mixtures (Lüscher *et al.*, 2014) that significantly increase aboveground biomass yield over monocultures (Finn *et al.*, 2013). Such benefit in mixtures can arise through positive species interactions (Nyfeler *et al.*, 2011) and complementary resource use (Hooper *et al.*, 2005), which suggests that high yielding mixtures transform high amounts of resources into biomass. High yield of grassland mixtures may thus suppress weed establishment through increased competition for resources (Frankow-Lindberg, 2012). Therefore, it can be hypothesized that weed suppression could be maximized through cropping targeted combinations of ley species in mixtures that ideally span a wide gradient of functional traits in order to optimize resource capture; this would leave a smaller share of resources to weeds.

Materials and methods

Five phytometer species (*Bromus hordeaceus* L., *Leontodon hispidus* L., *Poa trivialis* L., *Rumex obtusifolius* L., *Taraxacum officinale* Web.), representing common weeds of intensively managed grasslands, were grown from seeds and eight healthy seedlings per phytometer species were planted into ley stands in non-competing distances to each other in mid-May. Survival of phytometer plants was evaluated fourteen weeks after planting (beginning of September) by assessing their form and colour. Ley stands comprised four key forage species of productive temperate grassland: *Lolium perenne* L., *Cichorium intybus* L., *Trifolium repens* L., and *Trifolium pratense* L. Plots of 3×5 m were established following a simplex design (Cornell, 2002) with ley monocultures (2 replicates per species), binary mixtures (50% of each of two species, 2 replicates per pairwise combination), and a four-species mixture (25% of each of the four species, 3 replicates). Aboveground biomass of these leys was harvested six times per year; here, we report on the harvest after the evaluation of phytometer plants.

Five plant functional traits (PFT) related to resource acquisition and growth were measured on each ley species in each stand: leaf dry matter content, specific leaf area, leaf nitrogen (N) content, leaf carbon (C) concentration, and leaf C:N ratio. From these trait data, the functional diversity of ley stands was computed following Laliberté and Legendre (2010) using their index 'functional dispersion' (FDis). This index is zero by definition for monocultures and increases with increasing diversity of traits in mixtures. Finally, the survival of phytometers was regressed on functional dispersion of ley stands and their aboveground biomass yield using generalized linear mixed models, with survival of phytometer plants being a binary response variable (0 if dead, 1 if alive).

Results and discussion

Survival of phytometer plants was significantly suppressed both by higher functional dispersion of ley species (Figure 1, P=0.035 for the trend) and by higher aboveground biomass yield of ley stands (P<0.001, no figure shown). Specifically, phytometer survival was 48% in the functionally most diverse ley mixtures, and 65% in the functionally least diverse stands. Regarding the effect of biomass yield, calculated at mean FDis, phytometer survival was 34% in the most productive ley mixtures, and 58% in the least productive stands.

In parallel, FDis of the four-species mixture was distinctly greater than that of the binary mixtures (Figure 1, P=0.057, Wilcoxon rank sum test), while FDis of the binary mixtures was greater than that of monocultures (P<0.001). Moreover, the four-species mixture had 73% greater aboveground biomass yield than the average of monocultures (overyielding, P<0.001, test following Nyfeler *et al.* (2009)). Taken together, the targeted combination of ley species in mixtures led to higher functional dispersion

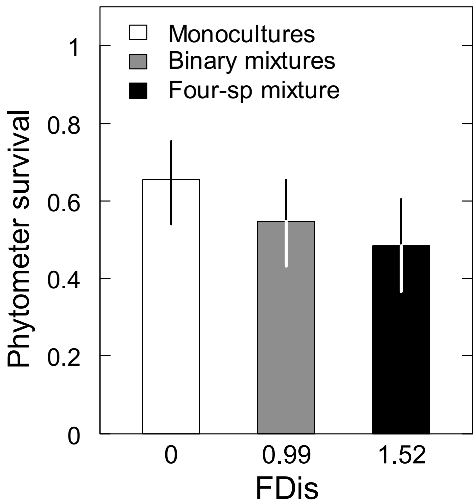

Figure 1. Phytometer survival for increasing values of functional dispersion (FDis), corresponding to monocultures, binary mixtures, and the four-species mixture (Four-sp mixture). Displayed is predicted phytometer survival ± standard error at mean aboveground biomass yield across all types of stands.

and aboveground biomass yield, both of which enhanced suppression of phytometer plants. This implies higher weed suppression through increased resource capture of leys because the measured PFTs are closely linked to resource uptake and growth.

The observed phytometer suppression could have been mediated through light interception effects (Sanderson *et al.*, 2012); yet, there is evidence that high yielding grassland monocultures and mixtures capture light to similar degrees (Frankow-Lindberg, 2012). Regarding belowground resources, ley mixtures take up more soil N and potassium than do monocultures (Hoekstra *et al.*, 2015; Nyfeler *et al.*, 2011), and increased soil N uptake can suppress weed biomass (Frankow-Lindberg, 2012). We thus argue that weed suppression can be enhanced by maximising pre-emption of and competition for multiple above- and belowground resources by the ley species. This could be achieved by (1) selecting specific ley species that have traits resulting in high resource capture and biomass yield, and (2) combining such ley species into mixtures of high functional diversity so that their complementary traits result in maximized total resource capture.

Conclusions

We conclude that increased acquisition of resources in functionally diverse ley mixtures enhances competition of the leys against weeds. Cropping such forage mixtures instead of monocultures can be a sustainable strategy to improve weed suppression.

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Assessment of soil structure and plant root patterns by means of X-ray micro computed tomography in grassland sites

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Abstract

Grassland sites are important ecosystems delivering essential ecosystem services such as carbon storage and filtering of water. Adequate management necessitates a better understanding of soil ecological processes in grassland ecosystems. Of special interest is the relationship between land use intensity and soil structure and their influence on root development. Soil structure, the spatial arrangement of pores and solid soil parts, is a key element in soil ecosystem functioning. The activity of soil biota, and the extent of plant root growth, combined with the nature of the soil type and its land use management practices will determine the kinds of soil aggregation patterns that will form. This field study was conducted to investigate the relationship between land use intensity and soil structure and their influence on root development in differently managed grassland sites. It is hypothesized that as the soil structuring, i.e. the solid soil surface/solid volume ratio increases, there would be a corresponding increase in the root surface and volume respectively. Undisturbed soil cores from 3 grassland plots from Swabian Alb 'Biodiversity exploratory' were investigated by means of X-ray micro-computed tomography. In the 3D images, the pore, solid and root phase could be clearly distinguished. Root surface correlated significantly with the estimated solid surface/volume. Preliminary results suggest a negative relationship between root development and land-use intensity.

Keywords: land use intensity, soil structure, roots, X-ray computed tomography

Introduction

Soil structure is affected by land use, cultivation intensity, rooting, abiotic soil properties, soil fauna, and climate. Management has a far-reaching influence on the soil system. Bronick and Lal (2005) provided an extensive review of the effect of management on soil structure in arable soils. Previous studies have shown that soil structure may depend on land use intensity. Thus, fertilizer applications may improve the soil structure (Haynes and Naidu, 1998), but they can also lead to a decrease of the soil organic carbon content and thereby reduce soil aggregation (Bronick and Lal, 2005). X-ray computed tomography (CT) is an imaging method with a 3-dimensional perspective which enables a quantitative assessment of soil samples in terms of soil structure and rooting pattern. Many studies have used this technique to investigate the impact of different fertilizer input and tillage practice (Munkholm *et al.,* 2013) on soil structure characteristics and to study their temporal dynamics (Garbout *et al.,* 2013). X-ray CT also enables a non-destructive investigation of the root system (Perret *et al.,* 2007). The overall aim of this field study is to understand the relationship between land use intensity and soil structure and their influence on root development in differently managed grassland sites.

Materials and methods

This field study was carried out in the Swabian Alb 'Biodiversity exploratory'. The overall experimental design as well as the site and climate conditions are described in detail by Fischer *et al.* (2010). The soil type of the three selected plots is classified as Leptosol whereas the land use intensities exhibit a wide range. Experimental site AEG05 is a fertilized mown pasture grazed by cattle and horses; AEG07 is not mineral fertilized but periodically grazed by sheep; and, AEG11 is a fertilized meadow without livestock. The selected sites were ranked in order from lowest to highest land use intensity; that is, AEG07, AEG11, and AEG05.

For sampling undisturbed soil cores, an automated sampling device was used to collect three undisturbed soil cores from the A-Horizon, with a size of 120 mm in height and diameter, followed by the extraction of smaller undisturbed soil cores with 30 mm in height and diameter (Kuka *et al.,* 2013b). The smaller soil cores were scanned by X-ray micro-computed tomography (µCT) with a resolution of 40 µm. In the 3D images, the pore, solid and root phase could be clearly distinguished and root/solid/pore volume as well as surfaces were estimated (Kuka *et al.,* 2013a).

Statistical analysis was conducted using the package PASW v.18.0. An error probability level of 0.05 was used throughout the tests as the threshold of significance.

Results and discussion

The largest root surface area, with mean value of 4,344 m^2 m^{-3} soil, was measured in plot AEG07. AEG05 had the lowest solid surfaces (mean value of 2,193 m^2 m^{-3} soil), whilst plot AEG11 was intermediate, with a mean value of 3387 m^2 m^{-3}soil. The differences between AEG05 and AEG11 versus AEG05 and AEG07 respectively were significant (t-test $P<0.05$). AEG07 had the highest root surface (mean value of 1,016 m^2 root surface m^{-3} soil) together with high percent root volume (mean value of 0.064 m^3 root volume m^{-3} soil); replicate AEG11c being similar in root volume (0.065 m^3 root volume m^{-3} soil) but not in root surface (581 m^2 root surface m^{-3} soil). The lowest values of root volume and surfaces were estimated in plot AEG05. With respect to root surface area, differences within sites were less than differences between sites. The standard deviation of estimated root surfaces within sites ranged between 5 and 66 m^2 root per m^3 soil volume. Significant differences existed between all the AEG plots in respect of the root surfaces (t-test $P<0.05$). These differences suggest that land use intensity is an important factor for root development (Kuka *et al.,* 2013a). Figure 1 shows the different rooting patterns with corresponding 2D X-ray µCT images of the three plots. On plot AEG05, sparsely occurring roots with coarse to fine diameter were observed. The solidified part of AEG05 is relatively high, suggesting compaction which at this site may have resulted from traffic by both agricultural machinery and grazing animals. The 2D X-ray µCT images of AEG11 show a fine-textured soil. This site is influenced by land machines but not by grazing. The roots of plot AEG11 appear overall to be of finer diameter than in the other plots and more evenly distributed than in plot AEG05. Due to landscape management, plot AEG07 is only sometimes grazed by sheep. The highly structured nature of this soil may be due to a combination of two possible factors: (1) the existence of relatively high content of carbonates typical for the site, and (2) low management intensity. The roots within the soil are distributed evenly and range in size from coarse to medium to very fine roots (Kuka *et al.,* 2013a).

Conclusions

As hypothesized, the root phase appears to be closely related to soil structure and strongly affected by land use intensity in differently managed grassland sites. Since soil structure heavily affects turnover processes in soil, this analysis may improve the understanding of relationships between biodiversity and ecosystem function in grassland soils.

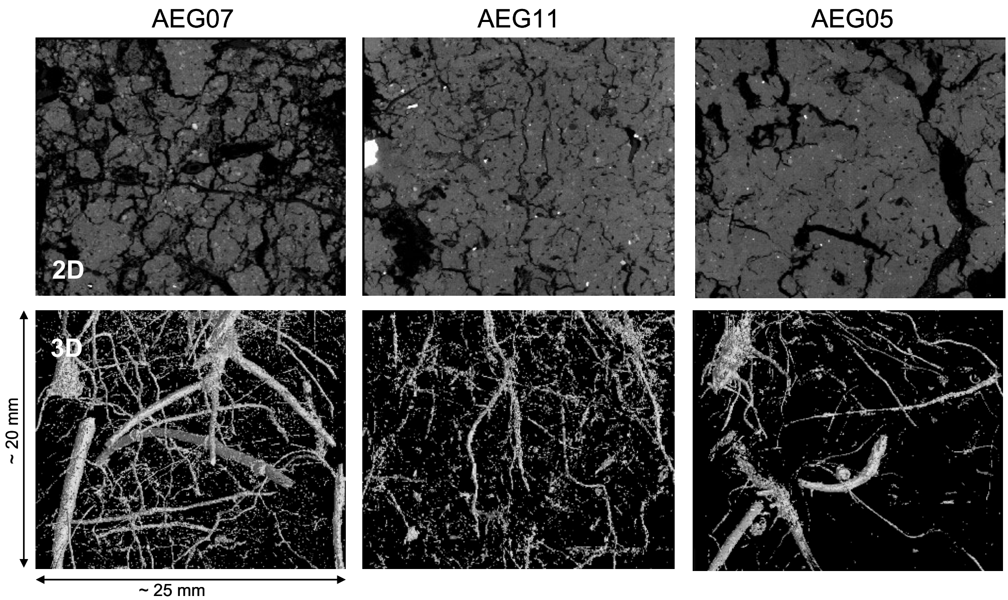

Figure 1. 2D X-ray micro computed tomography images and 3D segmented roots from plots AEG05, AEG07 and AEG11, ordered in or with increasing land use intensity.

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The influence of *Lupinus polyphyllus* Lindl. on energetic conversion parameters of plant biomass from semi-natural grasslands

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Abstract

European extensive grasslands have seen an increasing spread of the invasive alien plant species *Lupinus polyphyllus* Lindl., leading to a decline in biodiversity. Maintenance of biodiversity can be achieved by regular cutting before the ripening of the seeds of *L. polyphyllus*. The idea is to use the biomass for biofuel production. To investigate the effect of the lupine on energy conversion parameters, an artificial mixture series with silages of *L. polyphyllus* and semi-natural grassland was established, containing 0, 25, 50, 75 and 100% (fresh weight based) of *L. polyphyllus*. Mineral composition of silage and biofuel has been determined. It could be shown that lupine biomass is a suitable substrate for biofuel production when processed appropriately and that mineral concentrations in the solid fuel are appropriate for combustion.

Keywords: bioenergy, *Lupinus polyphyllus*, invasive plant, biodiversity

Introduction

Semi-natural grasslands are hotspots of biodiversity but are endangered due to cessation of management and intensification (Poschlod *et al.*, 2005). The success of invasive species threatens biodiversity additionally (Valtonen *et al.*, 2006). One of the invasive plant species in European grasslands is the legume *Lupinus polyphyllus* Lindl. (Lambdon *et al.*, 2008), which is indigenous to North America. To maintain and secure biodiversity in the multiple threatened semi-natural grasslands it is necessary to maintain a traditional management regime, with removal of biomass and nutrients (Jaquemyn *et al.*, 2003). In the case of *L. polyphyllus*, utilization of the biomass as feedstock is hindered through toxic alkaloids present in the plant species (Veen *et al.*, 1992). An alternative use for the biomass can be found in the energy recovery system investigated by the University of Kassel, the integrated generation of solid fuel and biogas from biomass (IFBB, Wachendorf *et al.,* 2009). The biomass is conserved as silage, followed by a hydrothermal conditioning step. After that a screw press separates the biomass into a press fluid (PF) rich in minerals (S, P, K, Mg, K, Cl) and sugars and a press cake (PC) rich in fibres. The PF can be used for biogas production and the PC can be used to produce a solid fuel. While the IFBB has been well investigated for semi-natural grassland biomass, there is no data available so far on the effect of plant biomass including *L. polyphyllus* on the feasibility of this energy recovery system. The aim of this study was to investigate the effect of *L. polyphyllus* on the produced solid fuel quality by producing an artificial mixture series of *L. polyphyllus* and extensive grassland silage with 0, 25, 50, 75 and 100% (fresh weight based) of *L. polyphyllus.*

Materials and methods

Two biomass types were collected in June 2014; grassland biomass without *L. polyphyllus* (L0) and pure *L. polyphyllus* biomass (L100). L0 was collected from the edge of a typical German lower mountain grassland area of the association 'Geranio sylvatici –Trisetetum' (R. Knapp ex Oberd. 1957). The L100 biomass was harvested manually. Both biomass types were left to wilt in open air overnight. After wilting they were separately chopped and ensiled in 30 l polyethylene barrels. A series of mixtures of both biomass types was ensiled in 3 replicates, containing 25, 50 and 75 weight % of *L. polyphyllus* and 75, 50 and 25 weight

% grassland biomass respectively (named L25, L50 and L75, respectively, from now on). After storing the barrels for 6 weeks, they were opened and the silage was mixed with water in a ratio of 1:4 (silage:water) in a concrete mixer with a maximum volume of 200 l. The mash was kept at constant temperature of 40 °C and stirred for 15 min. Subsequent mechanical dehydration of the silage was conducted with a screw press (type AV, Anhydro Ltd.). The conical screw had a pitch of 1:6 and a rotational speed of 6 revolutions min^{-1}. The cylindrical screen encapsulating the screw had a perforation of 1.5 mm.

Dry matter (DM) for silage, PC and PF was determined by oven drying at 105 °C for 48 h. Total ash concentration was determined by combustion in a muffle oven at 550 °C. The samples were analysed for C, H and N using an elemental analyser (Vario MAX CHN, Elementar Analysensysteme GmbH). S, K, P, Cl, Mg and Ca were analysed using x-ray fluorescence analysis. Mass flows of elements from the silage into PC and PF were calculated. Statistical analyses were done using the Software R. Analysis of variance with Tukey HSD as a post hoc test was performed to test for the effect of the proportion of *L. polyphyllus* on mass flows into PF within the IFBB system.

Results and discussion

Table 1 shows the DM, total ash and mineral concentrations of the silages and press cakes. For DM, the effect of *L. polyphyllus* is obvious, showing a very low concentration. The biomasses have high ash and mineral concentrations, making them unsuitable for the technical use as a solid fuel without pre-treatment. With the pre-treatment of IFBB the concentrations of minerals detrimental for combustion can be reduced in all cases, total ash concentration could be reduced from 9.1 to 5.1% DM for L100 and especially K, and Cl could be substantially reduced.

For all parameters investigated, the mass flows into PF were higher than the DM mass flow, which means their concentrations in the PC were reduced in comparison to the silage (Table 1 and 2). With the exception of ash, S and Cl, the mass flows were lower for the grassland biomass than for the *L. polyphyllus* biomass, indicating that the effect of mashing and dewatering was greater in the wet *L. polyphyllus* biomass than in the fibrous grassland biomass. The mass flows of the mixtures L25, L50 and L75 were intermediate compared to those of the two raw materials. The L0 biomass showed the lowest mass flow, whereas the mixtures and L100 were often not significantly different (Table 2). Even a minor addition of only 25 weight % of *L. polyphyllus* biomass to the grass biomass resulted in significantly increased mass flows for K, Mg and Ca.

Table 1. Arithmetic means of concentration of dry matter (DM), ash, N, S, K, Mg, Ca, Cl and P in the press cake (PC) in % for L0, L25, L50, L75 and L100.

		Silage					PC				
		L0	L25	L50	L75	L100	L0	L25	L50	L75	L100
DM	% FM	30.58	26.07	23.50	19.19	15.67	45.02	46.30	44.78	44.98	44.47
Ash	% DM	8.31	8.15	8.13	8.44	9.07	5.12	4.77	4.75	4.55	5.12
N	% DM	1.46	1.58	1.73	1.98	2.39	1.04	1.10	1.24	1.30	1.55
S	% DM	0.13	0.12	0.12	0.11	0.10	0.08	0.07	0.08	0.08	0.07
K	% DM	1.88	1.71	1.51	1.23	0.93	0.31	0.23	0.19	0.14	0.09
Mg	% DM	0.28	0.32	0.37	0.42	0.56	0.12	0.11	0.12	0.13	0.15
Ca	% DM	0.66	0.84	1.09	1.37	1.98	0.56	0.62	0.72	0.86	1.05
Cl	% DM	0.90	0.76	0.61	0.40	0.11	0.07	0.05	0.04	0.02	0.01
P	% DM	0.23	0.22	0.21	0.19	0.16	0.06	0.05	0.05	0.04	0.03

Table 2. Arithmetic means of mass flow of DM, ash, N, S, K, Mg, Ca, Cl and P into the PF with standard error of means for L0, L25, L50, L75 and L100.[1]

		L0	L25	L50	L75	L100
DM	%	15.6±1.5	25.9±5.5	25.4±4.5	25.4±4.3	31.5±3.2
Ash	%	48.0±1.2	56.6±3.4	56.5±2.3	59.8±1.9	61.2±4.9
N	%	39.5±1.2^{a}	48.4±4.2^{ab}	46.2±4.4^{ab}	51.1±2.0^{ab}	55.5±2.1^{b}
S	%	47.8±2.2	55.9±4.3	49.4±5.9	45.2±2.7	52.7±4.9
K	%	85.9±0.3^{a}	90.1±0.8^{b}	90.5±0.5^{b}	91.6±0.3^{bc}	93.3±0.4^{c}
Mg	%	63.7±1.1^{a}	73.8±1.6^{b}	75.0±1.7^{b}	76.9±0.9^{bc}	81.4±1.3^{c}
Ca	%	28.2±3.5^{a}	45.7±2.2^{b}	50.6±3.9^{bc}	53.3±2.2^{bc}	63.5±2.9^{c}
Cl	%	93.3±0.1	95.3±0.6	94.5±0.3	95.4±0.2	94.9±1.0
P	%	78.9±0.4^{a}	83.7±1.6^{ab}	82.2±1.3^{ab}	83.1±0.7^{ab}	86.2±1.0^{b}

[1] Uppercase letters indicate significant differences between groups.

Conclusions

Semi-natural grassland biomass with and without *L. polyphyllus* biomass has high mineral concentrations which hinder the thermal energetic use in combustion. However, with the IFBB pre-treatment, mineral concentrations could be substantially reduced. This reduction was higher in the case of N, K, Mg, Ca and P for the pure *L. polyphyllus* biomass than for the biomass from pure semi-natural grassland without *L. polyphyllus*. Even the addition of only 25 weight % of lupine biomass to semi-natural grassland biomass led to significantly higher reductions in the case of K, Mg and Ca. Thus, it can be concluded that biomass containing the invasive plant species does respond very well to the IFBB pre-treatment and delivers a solid fuel of at least the same quality than semi-natural grasslands.

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Valorising forage resources and conserving ecosystem services in marginal pastures

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Abstract

Marginal mountain pastures provide society with valuable products, unique biodiversity and appealing landscapes. Such ecosystem services are under threat from the abandonment of these areas and subsequent shrub encroachment. Our main aim was, therefore, to develop strategies for conserving biodiversity and productivity of subalpine pastures by low-input grazing with robust breeds. In part 1 of the project, we visited 19 sites across the Swiss Alps and carried out a botanical survey investigating the relationship between shrub cover and the composition of understorey vegetation. In part 2, a grazing experiment was carried out to study the biomass production of subalpine vegetation types as well as the growth performance of Dexter cattle and Engadine sheep. Part 1 revealed that the identity of the dominant shrub species has a strong impact on botanical composition. The understorey vegetation of *Alnus viridis* (green alder) was much more species-poor than that of *Pinus mugo* (creeping pine) and had a higher nutrient indicator value. Part 2 demonstrated that the understorey vegetation of *A. viridis* provides a largely underestimated forage resource for grazing animals. Together, these results indicate that conservation targets can be achieved by simultaneously producing meat from robust breeds.

Keywords: grazing, species richness, productivity, biomass

Introduction

Centuries of livestock grazing and human intervention have created mountain pastures of unique value to society. They provide forage for ruminants, habitat for numerous specialist species, protection against natural hazards and an attractive landscape for recreation and tourism. If grazing of mountain pastures declines or ceases, natural processes of succession change vegetation structure, habitat quality and ecosystem services. Throughout Europe, changes in mountain agriculture have led to the abandonment of marginal pasture areas and the subsequent expansion of woody plants. In the subalpine zone of the Swiss Alps, green alder (*Alnus viridis)* and creeping pine (*Pinus mugo)* dominate the succession on abandoned pastures (Brändli, 2010). The ongoing landscape transition has raised public concerns and there is a constitutional mandate to preserve the cultural landscape and the biodiversity. The aim of the project EG4BM (Extensive Grazing for Biodiversity and Meat production) was therefore to develop strategies for the conservation of biodiversity and the maintenance of productivity in subalpine pastures by low-input grazing with robust breeds.

Materials and methods

In the first part of the project (Part 1), we investigated plant species composition along 19 gradients from open grassland to closed shrubs, which were selected across the Swiss Alps in a stratified procedure. Along each gradient, plant species were recorded in 2 by 2 m quadrats at 0, 50 and 100% shrub cover. For all species, nutrient indicator values were extracted from Landolt (2010). Eleven gradients were dominated by *A. viridis* and six by *P. mugo*. Differences between the two shrub species at the different levels of shrub cover were tested using *t*-tests. In the second part (Part 2), we carried out a controlled grazing experiment at two sites situated on both sides of the Albula pass in Eastern Switzerland (46°34' N, 9°50' E). *A. viridis* was the dominant shrub species at both sites. Among other variables, standing biomass above 5 cm cutting height in the different vegetation types occurring in the grazing area was measured using exclusion cages of 1.2 by 1.2 m ground area. The data presented originates from 20 samples within the pasture area on the Eastern side of the pass.

Results and discussion

Part 1. The relationship between the occurrence of shrubs and the composition of understorey vegetation strongly depended on the dominant shrub species (Figure 1). Our data showed that the understorey vegetation of *A. viridis* was significantly less species-rich than that of *P. mugo*. At full cover of *A. viridis*, the understorey vegetation only consisted of a few specialist species. In contrast, vegetation at 50% cover of *P. mugo* was nearly as species-rich as vegetation at 0%. This evidence confirms indications of Pornaro *et al.* (2013) in the Southeastern Alps.

Higher levels of *A. virdis* cover were associated with elevated nutrient indicator values in the understorey plant community, which were significantly above those for sites dominated by *P. mugo*. This is likely due to the N_2 fixation of *A. viridis* in association with the bacterium *Frankia alni*. The additional N brought into this sensitive environment, in which plant growth is strongly limited by climate, increases the risk of N leaching and N_2O volatilization (Bühlmann *et al.*, 2014).

Part 2. Biomass production in the grazing experiment was clearly dependent on the vegetation type (Figure 2). At the site on the Eastern side of the pass, the understorey vegetation of full stands of *A. viridis* provided as much and sometimes more biomass than fertile pastures, which had comparably low productivity. Variation in understorey biomass was large and 95% of the measured values ranged between 0.7 and 2.9 t ha^{-1}, compared with 0.6 and 1.7 t ha^{-1} for the fertile pastures of the *Poion alpinae* association (Figure 2). Leaves of *A. viridis* are an additional biomass resource available to grazing animals and can amount to up to 3.8 t ha^{-1} (Wiedmer and Senn-Irlet, 2006).

The availability of abundant forage resulted in steady body weight gains of Engadine lambs and Dexter cattle, even in pastures where up to 80% of the area was covered by *A. viridis* (Zehnder *et al.*, 2016).

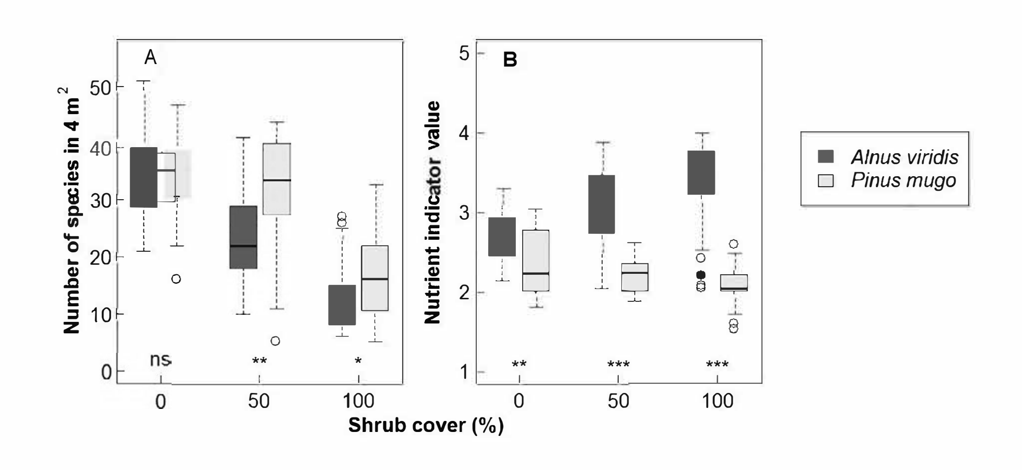

Figure 1. Relationship between the shrub cover of *Alnus viridis* and *Pinus mugo* and the average number of species in 2×2 m quadrats (A) and the community-weighted mean of the nutrient indicator value (B) of the understorey vegetation. Bold lines show median values, boxes the 50% quantile range and whiskers 1.5 of the interquartile range. Significance of *t*-tests between the two dominant shrub species are given as *** $P<0.001$, ** $P<0.01$, * $P<0.05$ and ns non-significant.

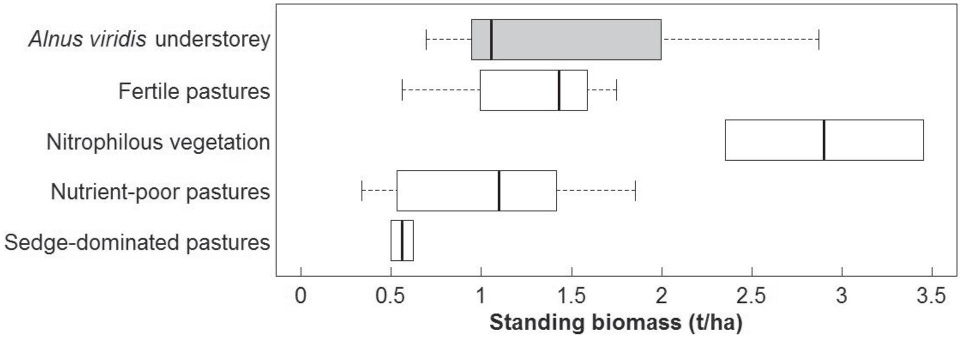

Figure 2. Standing biomass above 5 cm cutting height in t ha^{-1} in five vegetation groups measured at one site of the grazing experiment (2,000-2,200 m a.s.l.). Bold lines show median values, boxes the 50% quantile range and whiskers 1.5 of the interquartile range.

Weight gain of Engadine lambs tended even to be higher on shrub-encroached than on open pastures, possibly because this sheep breed has a preference for *A. viridis* leaves and understorey vegetation.

Conclusions

Our investigations demonstrated that the composition of understorey vegetation is strongly associated with the identity of the dominant shrub species. *A. viridis* turned out to be a serious threat to biodiversity, probably due to a combination of shading and eutrophication of the environment. On the other hand, the understorey vegetation of *A. viridis* provides a largely underestimated forage resource for grazing animals. These results indicate that conservation targets can basically be achieved whilst simultaneously producing meat from robust breeds.

Acknowledgements

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The first pillar of the new CAP – implications for low input grasslands

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Abstract

The first drafts of the CAP (Common Agricultural Policy) for the period 2014 to 2020 strived to give more emphasis and support to extensive grasslands in Europe. However, the final outcome is a highly complex mix of old and new rules containing many loopholes. In consequence, Member States (MS) can continue a more or less business as usual strategy, although the administrative burden has risen. However MS intending to support farms according to their provision of environmental services are thwarted by the recent regulations. This is the case, in particular, for extensive grasslands. The new 'greening' measures imply only a small stimulus for high nature value farming in general. This particularly applies to species-rich grasslands. Aspects such as the broader definition of the eligible area appeared promising at first sight. Unfortunately, the way these definitions were refined in the implementing legislation substantially limits their potential positive impact. This paper discusses aspects about how the 1st pillar of the new CAP is affecting/influencing the use of extensive grasslands and their inherent biodiversity throughout Europe.

Keywords: European Union, common agricultural policy, first pillar, extensive grasslands

Introduction

In the previous two decades the support mechanisms of the CAP were gradually reformed. Starting from a nearly pure price support system in the early 1990s, the CAP first shifted to the so-called coupled payments (linkage of payments to cropped area or livestock unit). Recently, area-based payments have been the main support mechanism of the CAP. In theory, this support is supposed to have no impact on the amount of agricultural production in the EU, as it is granted even if the area is only mulched.

The CAP reform of 2014 followed the path set by the so-called Fischler reform of 2003. The recent reform entails a movement to more uniform distribution of the payments per hectare across the EU, a slightly stronger emphasis on environmental issues, and a higher degree of freedom for Member States (MS) and regions regarding the financial allocation of the CAP funds. Taking into account all subsidies, public support derived from CAP instruments accounts for 40% of the average agricultural income across the EU (EC, 2015a, 2016). In particular, farms in marginal areas strongly depend upon CAP payments and can thus be heavily affected by the recent changes in the support system (EC, 2015b,c).

Furthermore the CAP defines standards that must be met by farmers in order to receive grant money. These standards cover administrative issues such as ear tags for livestock, or the precision with which the location and shape of an agricultural parcel is registered, but also the composition of sward, animal welfare and environmental issues.

This paper tries to assess the implications the 2014 CAP reform for the preservation and management of species rich grasslands ecosystems in the EU.

Materials and methods

We base our assessment on an analysis of the legal texts supported by a literature review of ex-ante assessments. We focus on the compulsory decoupling, the shift from farm-specific historical payments towards more regionally-generalized payment schemes, the delineation and delimitation of the eligible area and the implications of the so-called Greening of the 1st Pillar of the CAP.

Results and discussion

Uniform area payments for cropping and grasslands

By 2020, all EU MS have to adjust their payment systems from farm-specific historical payments to a system that is at least partially based on uniform regional payments per hectare. 21 MS decided to implement a flat rate payment for the entire state by 2020 at the latest. This is a logical step away from production-linked payments which had the side-effect of discriminating low input grasslands. In general, low productivity systems such as suckler cow farming and sheep breeding will benefit. The rationale is that the stocking rate in these systems is sufficiently low that the loss of payments linked to historical herd size is overcompensated by the increasing area payments (Offermann *et al.,* 2016). However, without animal-related payments, the gross margins for suckler cows and ewes are frequently negative (Deblitz, 2015). In particular in marginal areas, this might lead to a loss of livestock and a further extensification and possible abandonment. This has potentially negative impacts on the biodiversity of such extensive grazing systems (Luick *et al.*, 2015). The possibility of significant declines of the livestock herd in marginal regions may be the reason why 25 MS will continue to grant animal payments for suckler cows and/or ewes in the CAP's 1st pillar (EC, 2015d).

Determination and delimitation of eligible area

The higher the economic importance of area-related payments in relation to the payments per head, the more important is the correct determination and delimitation of the eligible area for grant money. The positive effect of a flat-rate payment system for low input farm/grassland system only holds if a substantial share of the grazed area is eligible for payment. In order to understand this effect, the cornerstones of the CAP must be understood: the CAP's 1st pillar payment should be a simple system to administer on a European level, as well as allowing unambiguous cross audits. One requirement for eligibility for payment is the linkage of the area to agricultural production (at least in the recent past). By definition, this implies that areas without agricultural production, e.g. rock outcrops and wide hedges, do not belong to the area eligible for direct payments. While it is fairly easy to determine the size and shape of cropped farmland and intensively managed grasslands, the issue becomes trickier when applied to pastoral systems used at low intensities. Often these systems are characterised by gradients rather than sharp boundaries between the different land covers, e.g. wooded areas, thickets, wetlands, grasslands, heath and barren areas. Furthermore, these different land covers are often intimately interwoven and their extent can fluctuate both within and between years. A land classification system that is not in accordance with the legal rules set by the EU can have significant financial consequences both for farmers and MS. However, the CAP's requirement to classify traditional, large-scale pastoral grasslands unambiguously is not feasible, or at least it cannot be achieved within the predefined margins of error.

Definition of grassland eligible for CAP payments

With the recent CAP, the definition of grassland eligible for direct payments has changed: MS can now opt to include areas that 'are grazed and not predominantly covered by grasses and herbs'. Furthermore, MS can extend this area by defining habitat types depending on agricultural use. However, only the area covered with forage plants in these habitat types is eligible for CAP 1st pillar payments. Especially in low productive ecosystems, e.g. highly endangered habitat types such as *Corynephoretum canescentis*

-grassland, mosses or bare soil can cover more than 50% of the ground. However, the area is only eligible if more than 50% of the surface is covered by edible plants. Likewise, traditionally mown systems are still not included. This means that mown reed stands (*Phragmites australis)* or wet meadows, with a high share of sedges, are not eligible for CAP 1st pillar payments.

Greening of CAP 1st pillar payments

The obligation to maintain permanent grassland replaced a similar regulation previously included in CC. The minimum requirement of the EU for the MS, on a regional level, is that the proportion of farmland which is grassland must not decline by more than 5% between 2012 and 2020. Only in very few regions have higher rates of conversion occurred in the recent past. A new element is that farmers must not plough (in the sense of destroying the sward) 'environmental sensitive permanent grassland'. In most regions this grassland is defined as a certain share (up to 100%) of the grassland in Natura 2000 areas (EC, 2015d). However, compared to the other elements of the Greening (crop diversification and provision of ecological focus areas (EFA)) the penalties for violating the obligations for grassland protection are modest. It can therefore be advantageous for a farmer to convert grassland to arable land, accept the penalty and then establish an EFA after one year of cropping. Such an EFA can even be an area seeded with intensively managed clover.

Conclusions

The CAP reform 2014 strengthens low input grassland cultivation. However, it is unlikely that the level of support is sufficient to reverse the overall decline of these systems. Instead, the CAP Greening rules could even act as an incentive for the conversion of grassland. The mid-term evaluation of the multiannual financial framework in winter 2016/17 and the European Commission's report on Greening in 2017 may induce major changes of the CAP even before the scheduled reform in 2020.

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Soil acidity effect on agrocenoses with perennial grasses

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Abstract

To determine the influence of soil acidity on growth of perennial grasses and soil biological properties, experiments were performed with variable soil acidity: naturally highly acidic (pH_{KCl} 3.9) and medium acidic (pH_{KCl} 5.0). Four perennial grasses mixtures were cultivated. The mixtures where composed of one variety of legume and two grasses: *Trifolium pratense* L., *Trifolium repens* L., *Trifolium hybridum* L., *Medicago sativa* L. (each of 50%) with *Phleum pratense* L. (35%) and *Poa pratensis* L. (15%). The aim of this study was to assess the impact of soil acidity on different perennial grass mixtures' growth and development, and on soil biological properties. Root distribution in the soil profile was determined by the plant species. The negative effect of highly acidic soil was shown on the mixture with *Trifolium hybridum* L., where the amount of roots at 0-10 cm soil depth was lower by 44.2% (P<0.01) in the second year of development. In the mixture with *Trifolium repens* L., roots developed better in highly acidic soil; the total amount of roots was 2.5 (P<0.01) times higher than in medium acidic soil. Although differences were found in root development, the total aboveground mass of perennial grass mixtures showed better growth in medium acidic soil.

Keywords: soil pH, perennial grasses, root biomass, total microorganism biomass, CO_2, earthworms

Introduction

Ecosystem stability and its functions are greatly influenced by anthropogenic factors. The biodiversity of sustainable grassland systems is a key to mitigating the impacts on soil biota influenced by soil management. It is important to understand the influence of soil and plant interactions through plant/roots, soil biota and soil properties on ecosystem functions (Bell *et al.*, 2014; Van Eekeren *et al.*, 2007). Perennial grasses in agrophytocenoses create a specific microclimate, roots are strongly involved in sward formation, they influence soil structure, increase porosity, improve aeration, break down minerals, whilst root exudates and litter from roots and aboveground parts supply nutrients to the soil food web (Van Eekeren *et al.*, 2007). Of ecological significance are root allocation and distribution, as well as their total length and surface area of roots by which water and nutrients are absorbed. Soil pH has great influence on plant nutrient uptake, yield and survival, as very acid conditions lead to unsuitable forms of mineral elements (Repšienė and Skuodienė, 2010). The aim of this study was to determine the influence of soil acidity on growth of perennial grasses and soil biological activity.

Materials and methods

In the Vezaiciai Branch of the Lithuanian Research Centre for Agriculture and Forestry experiments were performed to determine the influence of soil acidity on the growth and development of perennial grass mixtures with legumes and on soil biological activity. The soil of the experimental site was (*JIn) Dystric Albeluvisols (ABd*), formed on medium-moraine loam. Soil was limed with dolomite once in the autumn of 2013. Different pH_{KCl} levels of the soil were formed: highly acidic (pH 3.9) and medium acidic (pH 5.0). Soil agrochemical characteristics were determined from the depths of 0-10 and 10-20 cm. Available aluminium was estimated by Sokolov's method. P_2O_5 and K_2O were determined by the A-L method, total nitrogen by Kjeldahl. In the spring of 2014, *Hordeum vulgare* L. was undersown with four different perennial grass mixtures with legumes: *Trifolium pratense* L. (TP), *Trifolium repens* L. (TR), *Trifolium hybridum* L. (TH), *Medicago sativa* L. (MS) (each of 50%), combined with *Phleum pratense* L. (35%) and *Poa pratensis* L. (15%). Experimental plots were arranged in randomized block design with four replications. At the end of the growing season, plant root mass was determined from the soil layers of 0-10 and 10-20 cm by the Katchinski monolith washing method. Phythomass was recalculated into dry mass. Total nitrogen was determined by the Kjeldahl method, organic carbon by mineralizer 'Heraeus'. Earthworm abundance and biomass was measured through the excavation of a 25×25×20 cm blocks, and through hand sorting. Soil CO_2 emission was determined by titration method and total biomass of microorganisms – by fumigation method. Statistical analysis was carried out using ANOVA. Soil agrochemical characteristics from the year 2014 are presented in Table 1.

Results and discussion

The development of perennial grass mixtures depended essentially on soil pH and biological properties of the plant species that formed the phytocenoses. In the first year of development plant root mass was essentially greater in soil with pH 3.9 (Table 2). In the second year of grass development these differences levelled off and because of favourable conditions for plant nutrition. Root mass in soil with pH 5.0 was then slightly higher (by 12%) than in soil with pH 3.9. Regardless of the pH of the soil, the greatest root mass in the soil depth of 0-10 cm was found with the mixture with red clover. At the depth of 10-20 cm in medium acidic soil, the alfalfa mixture developed greatest root mass.

The belowground phytomass C:N ratio changes differed little between the plant mixtures. The difference in soil pH had a marked influence on soil biological activity. According to the release of CO_2 from the soil, the respiration and mineralization processes and microbial activity were essentially higher in soil with pH 5.0 than with pH 3.9. Due to increased activity, the total microbial biomass was significantly

Table 1. Soil agrochemical characteristics at experiment establishment in 2014.

Variable	Soil pH 3.9		Soil pH 5.0	
	0-10 cm	10-20 cm	0-10 cm	10-20 cm
Available Al, mg kg^{-1}	88.59±11.73	89.44±12.58	3.88±2.22	43.27±6.95
Available P_2O_5, mg kg^{-1}	162.63±10.33	161.88±8.88	135.31±6.49	135.44±6.96
Available K_2O, mg kg^{-1}	201.38±5.68	200.38±3.80	195.50±6.69	192.00±5.83
N_{Total}	0.13±0.00	0.13±0.00	0.13±0.00	0.14±0.00

Table 2. Effects of pH-regime on morphological and biological parameters of grass mixtures and some soil biological properties in the first year of development.[1,2]

Soil pH	Grass mixtures	Roots 0-10 cm		CO_2 (mg g^{-1})	MO biomass (µg C g^{-1})	Earthworms	
		mass (g m^{-2})	C:N			amount	biomass
pH 5.0	TP	62.3	25	0.046	135.22	16	13.0
	TR	54.5	22	0.039	130.11	16	12.1
	TH	45.5	20	0.050	116.11*	18	10.6
	MS	55.6	24	0.038	117.00	19	12.9
pH 3.9	TP	125.2**	28	0.023**	147.44	9	3.2*
	TR	140.9**	26	0.027**	133.78	6	1.4*
	TH	149.2**	21	0.024**	160.22**	12	5.2
	MS	107.6	23	0.024**	127.89	13	5.4
	LSD_{05}	51.5	11	0.008	18.30	12	9.4

[1] TP = *Trifolium pratense*; TH = *Trifolium hydridum*; TR = *Trifolium repens*; MS = *Medicago sativa*; LSD = least significant difference.

[2] * and ** indicates significance at $P<0.05$ and $P<0.001$.

lower than in soil pH 3.9, especially with hybrid clover (in the first and second year of development) and alfalfa mixtures (in the second year of development).

The evaluation of earthworm population abundance indicated that conditions were more favourable for earthworms in soil with a pH of 5.0; in the first and second years of development, earthworm amount was higher by 70 and 81%, respectively, than in highly acidic soil. In the second year of development, following the increment in root mass, the number of earthworms increased as well (Tables 2 and 3).

Although differences were found in root development, the total aboveground mass of grass mixtures showed better growth in soil with pH 5.0, resulting in an average dry mass yield of 9.36 t ha^{-1}, significantly higher than in soil with pH 3.9 (6.80 t ha^{-1}) (LSD_{05} =1.87).

Table 3. Effects of pH-regime on morphological and biological parameters of grasses mixtures and some soil biological properties in the second year of development.[1,2]

Soil pH	Grass mixtures	Root mass		CO_2 (mg g^{-1})		MO biomass (µg C g^{-1})		Earthworms	
		0-10 cm	10-20 cm	0-10 cm	10-20 cm	0-10 cm	10-20 cm	amount	biomass
pH 5.0	TP	659.2	28.9	0.044	0.007	230.89	234.56	23	7.6
	TR	155.6**	29.1	0.041	0.009	201.67*	247.67	32	12.4
	TH	513.6	26.8	0.043	0.013	191.89**	215.44	33	12.5
	MS	545.6	76.2	0.043	0.012	160.22**	175.33**	27	11.2
pH 3.9	TP	557.5	37.4	0.030*	0.012	323.89**	397.67**	11	3.1
	TR	394.1	30.4	0.034	0.009	327.00**	379.11**	19	10.3
	TH	286.4**	22.5	0.026*	0.010	306.56**	403.22**	21	10.2
	MS	460.5	27.8	0.028*	0.008	283.00**	340.11**	11	2.4
	LSD_{05}	294.7	48.2	0.014	0.009	28.61	22.98	14	10.0

[1] TP = *Trifolium pratense*; TH = *Trifolium hydridum*; TR = *Trifolium repens*; MS = *Medicago sativa*; LSD = least significant difference.
[2] * and ** indicates significance at P<0.05 and P<0.001.

Conclusions

Based on the results of this experiment, it can be concluded that the root mass and distribution among soil layers in the first year of development depended on the soil pH, whilst in the second year of development root mass was influenced by the grass species. The difference in soil pH significantly affects the soil biological activity. In soil with pH 5.0, earthworm abundance, soil respiration and microbial activity were essentially higher, and microbial biomass was lower, than in soil with pH 3.9.

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Mixtures provided similar benefits to nitrogen yield under grazing and under mowing

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Abstract

Previous studies showed benefits of mixing species with complementary characteristics related to nitrogen (N) capture on phytomass and N yields of mown grassland. Grazing instead of mowing may impact soil properties and the interactions between plant species. This study aimed at determining whether grazing modifies the benefits of mixtures on N yield compared to mowing. The design included N_2 fixing and non-fixing species, as well as shallow- and deep-rooting species. *Lolium perenne* (Lp) monoculture and mixtures with *Cichorium intybus* (Ci), or/and *Trifolium repens* (Tr) and *Trifolium pratense* (Tp) were compared under grazing or mowing for their N yield and capture of fertilizer and atmospheric N. Mixtures of the N_2 fixing and the non-fixing species yielded as much N as the *L. perenne* monoculture fertilized with 2.4 times more N, showing the tremendous benefit of mixtures for N efficiency. The addition of the deep-rooting *C. intybus* did not lead to a significantly larger N capture. The benefits of the mixtures on N yield were similar under grazing and mowing. Grazing did not modify the proportion of N derived from fertilizer and symbiotic fixation in the plants. We conclude that grass-legume mixtures are effective in improving N efficiency under both mowing and grazing.

Keywords: multispecies swards, mixture effect, nitrogen yield, grazing

Introduction

Recent studies showed that mixing grasses and legumes clearly benefits phytomass and nitrogen (N) yields in fertilized swards managed under mowing (Suter *et al.*, 2015). Complementarity between N_2 fixing and non-fixing species (Nyfeler *et al.*, 2011), as well as temporal and vertical niche differentiation are processes involved in the mixture effects benefiting productivity and nutrient capture of mixtures (McKane *et al.*, 1990). But grazing may modify these processes as compared to mowing by its effects on plant growth, nutrient availability and spatial redistribution in the soil, as well as on soil compaction (Bilotta *et al.*, 2007). The effects of mixing grass, forb and legume species on nutrient capture might therefore differ markedly under grazing and mowing. On the other hand, data from a previous study indicated that the relative advantage for phytomass production of a four-species grass-legume mixture, as compared to a *L. perenne* monoculture, was similar when grazed and when cut (Collins *et al.*, 2014). The objective of our study was to determine whether grazing modifies the benefits of multispecies swards on N yield as compared to mowing.

Materials and methods

Rotational grazing and mowing (management types) were compared on five sward types in a split-plot design with 3 replicates, with the sward type as main plot and the management type as subplot. This design is especially powerful for testing the interaction between management type and sward type. The 453 m^2-large pasture plots were grazed 6 times per year with 2 heifers per plot. The subplots under mowing were cut at the start of each grazing period. The five sward types were: (1) monoculture of *Lolium perenne* (Lp) (MonoLp), (2) mixture of Lp and *Cichorium intybus* (Ci) (LpCi, 2/3 Lp + 1/3 Ci), (3) mixture of Lp, *Trifolium repens* (Tr) and *Trifolium pratense* (Tp) (LpTrTp, 2/3 Lp + 1/6 Tr + 1/6 Tp), (4) mixture of Lp, Ci, Tr and Tp (LpCiTrTp, 1/2 Lp + 1/6 of each of the other species), and (5) monoculture of Lp fertilized with 2.4 times more N than the other sward types (MonoLp_HN, fertilization of 350 kg N ha^{-1} $year^{-1}$). The four species were selected for their differences in N_2 fixing ability and rooting depth (non-fixing: Lp and Ci, fixing: Tr and Tp, shallow-rooting: Lp and Tr, deep-rooting Ci and Tp). In the grazed plots, the yield was estimated by measuring the height of the swards before and after grazing with a rising plate meter (83 measurements per plot). The N derived from fertilizer application (Nfert) and symbiotic N_2 fixation in Tr and Tp (Nfix) were measured using ^{15}N-labelled fertilizer applied on subplots (e.g. Nyfeler *et al.*, 2011). The N derived from the soil (Nsoil) was calculated as the difference between the total N content and the sum of Nfix and Nfert. The 3 years total dry matter and N yield data were analysed by ANOVA.

Results and discussion

The dry matter yields of the LpTrTp and the LpCiTrTp mixtures were significantly larger than those of the MonoLp sward (+47% on average over the grazing and mowing management; Table 1). This difference was +78% for the total N yield. These two mixtures even equalled the yield of the highly fertilized *L. perenne* monoculture (MonoLp_HN). This confirms the tremendous benefit of grass-legume mixtures as compared to grass monocultures in terms of N efficiency. The interaction between the sward and the management types was not significant for either dry matter or total N yield (Table 1), which shows that the benefit of the mixtures on dry matter and N yield was similar under grazing and mowing. The addition of the deep-rooting *C. intybus* in the swards did not lead to a significantly larger N capture (LpCi versus MonoLp, and LpCiTrTp versus LpTrTp), indicating that the observed mixture effect on N yield was mainly due to symbiotic N_2 fixation and not to vertical niche differences in N uptake from the soil.

In each of the four species of the LpCiTrTp mixture, the total N content in the harvested phytomass were similar under grazing and mowing (P=0.89), as well as its components (Nfert, Nsoil and Nfix; Figure 1). Thus, although grazing induced compaction of the upper soil layer (0-6 cm soil depth; data not shown), it did not modify the ratio of N derived from symbiotic fixation, fertilizer or the soil in the different species. The absence of a negative effect of grazing on symbiotic N_2 fixation by legumes probably contributed to the similar mixture effects under the two managements.

Table 1. Effect of the sward types on the dry matter and the total nitrogen yield.[1]

Sward type	N fertilisation (kg N ha^{-1} yr^{-1})	Dry matter yield (kg ha^{-1} yr^{-1})	Total N yield (kg ha^{-1} yr^{-1})
Monoculture MonoLp	145	4.7a	118a
Mixture LpCi	145	5.7b	142a
Mixture LpTrTp	145	6.8c	206b
Mixture LpCiTrTp	145	7.0c	213b
Monoculture MonoLp_HN	350	6.7c	225b
Standard error of the mean		0.4	13
P-value ANOVA			
Sward type		<0.05	<0.01
Management type		<0.01	<0.01
Interaction		0.88	0.67

[1] The data are the average of 3 experimental years. The average of both management types is shown, as the interaction between sward and management types is not significant. Means followed by a common letter are not different at the 5% level (Duncan MRT).

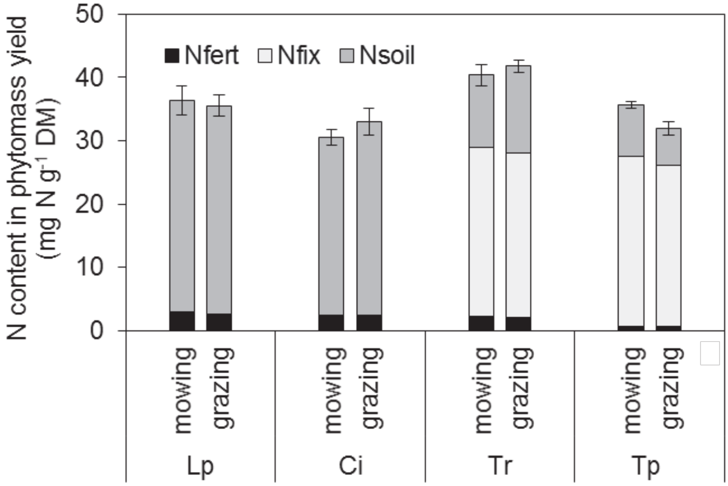

Figure 1. Nitrogen content in the harvested phytomass of each species of the mixture with *Lolium perenne* (Lp), *Cichorium intybus* (Ci), *Trifolium repens* (Tr) and *Trifolium pratense* (Tp) managed under mowing or grazing. Total N content is divided in N derived from the fertilizer (Nfert), N derived from N_2 fixation (Nfix) and N derived from the soil (Nsoil). The data show the mean of two regrowths and the error bars the standard error of the mean.

Conclusions

As compared to a *L. perenne* monoculture, the benefits of multispecies swards on phytomass and N yield were equally impressive under grazing and mowing. We conclude that grass-legume mixtures are effective for improving N efficiency under both mowing and grazing.

Acknowledgements

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Maintaining grasslands on cattle farms: the role of local social dynamics

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Abstract

For at least ten years now, grasslands have been experiencing a revival of interest, due to their multiple functions: grasslands in the forage system limit the cost of animal feed; they are an asset in the quality of products issued from pastures; they play numerous roles regarding biodiversity, ecosystemic functions, and landscape. Nevertheless, increasing the part of grasslands in crop rotation is a real challenge in France, where the modernization of agriculture has continually driven to its lowering. However, in cattle breeding territories where grasslands are globally decreasing, some areas keep a notably more important part of grasslands; our study aims to understand why. We chose the area of Boulonnais (France), as field for a sociological investigation within a national research program. Qualitative interviews were led with various local stakeholders to understand which factors can explain such a persistence of grasslands. Results show that beyond natural factors such as suitable climate and steep slopes, where arable crops are impossible to grow, the management of the Common Agricultural Policy agrienvironmental measures by the Regional Natural Park Caps et Marais d'Opale relayed the action of agricultural extension services, and brought new expectations towards grasslands.

Keywords: grasslands, social dynamics, low grassland territories, Boulonnais

Introduction

After more than fifty years of intensive dairy or beef cattle breeding, where farming systems developed on the 'maize silage-Italian ray-grass' model, except in some disadvantaged regions, grasslands have been experiencing a revival of interest these last thirty years. Fodder systems including more grass leys appeared, and in regions with temperate climates, the association of English ray-grass and white clover extended (Huyghe and Delaby, 2013). This change is also due to their multiple functions: grasslands in the forage system limit the cost of animal feed; they are an asset in the quality of products issued from pastures; they play numerous roles regarding biodiversity, ecosystemic functions, and landscape (Sabatier *et al.*, 2015). However, despite the existence of agronomic and technical innovations, and the 'greening' of the Common Agricultural Policy, the orientation choices of the professional structures over the past thirty years have contributed to the global decline of the share of grassland in crop rotations. In our national research program, the focus was set on breeding territories showing an overall decrease of grasslands (a loss of 2% between 2000 and 2010 (Agreste, 2010)). Within these territories, some areas maintain a notably more important part of grasslands and do not follow the overall dynamics (Couvreur *et al*, 2016). Such a paradox led us to wonder about the determinants of this specificity, and to assume that social dynamics – i.e. interactions between individuals – could explain the local maintenance of grasslands.

Materials and methods

The studied area is located in the area of Boulonnais, to the north of France. This canton has a large part of grassland in the total Utilized Agricultural Area, and also shows a low decrease of grass surfaces in comparison with the regional trends. The presence of breeding – 75% of all farms are specialized in dairy farming – makes it an interesting zone. The area is characterized by a dense river network, a very steep relief, and hydromorphic calcareous soils. The maritime climate, cold and often wet, does not always enable crop maturity, but is favourable to meadows, since grass is a good way to value the 1,200 mm annual rainfall. The area is part of the Regional Natural Park (RNP) of Cap et Marais d'Opale, which gathers 152 towns on 130,000 hectares. A RNP is a label given by the French state to a rural territory. This territory brings together towns whose landscapes, natural habitats and cultural heritage are of high quality, but may also show weaknesses. The objective of the RNP is therefore to protect and enhance this remarkable local heritage, while maintaining social and economic activity, including agriculture, which here covers 70% of the territory.

The choice was made to collect qualitative data by individual semi-directive interviews with local actors, likely to influence the spatiotemporal evolution of grasslands. Seventeen actors were met in December 2013: six farmers were representatives of the local cooperatives' equipment sharing scheme, of the farm unions, or of a local farmers group. The other persons were representatives of the Regional Natural Park (one), of a dairy company (one), of a local cooperative (one), of the local communities (three), or of the agricultural extension services (three) or researchers (two). The questions were focusing on land use and climate, farm systems, perception and expectations about grasslands, and on the future of the area. The interviews were recorded and transcribed, so as to later carry out content analysis, allowing us to rank the different opinions in themes.

Results

The presence of grasslands in this canton can be explained both by natural and social factors.

The farming community – either farmers or agricultural advisors – tends to mention the natural factors as constraints. They see the steep slopes, the hydromorphic character of soils and the climate as factors which make the use of these lands for arable crops almost impossible. They share the idea that the area is primarily devoted to cattle breeding, based on the use of grass forage. Besides, grass can limit erosion and runoff on the steep slopes. They add the fact that sometimes some plots are hard to reach with farm machinery, and that ploughing permanent grasslands is prohibited. Yet, in their opinions, grass is more a forced choice than an asset to value. Their judgments are based on a short term economic logic, where grass is systematically compared to cereals, whose prices are more profitable.

Interviewees, whose jobs or functions are not directly in relation with agriculture, consider grasslands differently. They emphasize the critical role the Regional Natural Park plays in maintaining grasslands. Actually, the RNP decided to support and to promote sustainable agriculture. Thus, using the European Common Agricultural Policy, the RNP proposed farmers to subscribe to agri-environment measures. The RNP even proposed a specific measure entitled 'Maintaining the Boulonnais hedgerow' for the restoration of ponds, the plantation of hedges and for maintaining grasslands. Its overall goal is to preserve water quality, to prevent erosion and landscape degradation, and to enhance biodiversity. Preserving water quality was indeed a priority for Boulonnais between 2007 and 2013, because the nitrate concentration in the water was exceeding 50 mg l^{-1}, placing it among the 'vulnerable zones'. This measure was complementing the national agri-environment measures, among which the 'grassland premium' encourages the management of extensive grazing, i.e. below 1.4 Livestock Unit ha^{-1}. Moreover, the RNP implemented a grass management training program for farmers, with the help of the local Chamber of Agriculture, of the French Association for seeds and seedlings (GNIS), and of a local farmers group (GRDA). Different themes were discussed such as meadow diagnosis, pasture management, choice of the species and varieties, and creation of fodder collections. Project studies were led about barn drying systems, to secure the production of quality hay. The RNP also helped the local farmers group to buy a seed drill in common. Finally, in 2013, the Park organized a day seminar on grasslands in relation with livestock farmers' activities. All these actions were carried out with the idea of preserving ecological resources. The results of the survey show that the dynamism of the RNP is unanimously appreciated. As one of the interviewees notices: 'Ten years ago, grasslands were not considered as real crops. Today the mentalities have changed, thanks to support and information provided'. Moreover, maintaining grasslands is congruent with organic agriculture, which is already quite common in this area, compared to the North department as a whole. Since organic products get higher prices, the RNP encourages it, as another design for viable farm systems, less dependent on European subsidies. Some interviewees are even wondering whether they should implement a territorial quality sign for feed products coming from farming systems based on grass. Therefore this shows that with a multifunctional approach, new projects can emerge.

Discussion

As observed in Lieue de Grève in Brittany, or in the Vittel area in Vosges (Perrot-Maitre, 2006), maintaining grasslands is encouraged for environmental purposes. The need to preserve water quality can be the main factor that decides a non-agricultural organization to enforce a territorial policy addressed to farmers, whether it be a RNP, a community of towns, or a private company selling mineral water. It highlights the fact that grasslands have territorial functions, beyond their strictly agro-zootechnical interest, that lead non-agricultural organizations to interfere with the agricultural development. In a context where the French government emphasizes the need for the agricultural sector to contribute to climate change mitigation, and where consumers' demand for organic feed is increasing, grasslands could be the cornerstone of an agro-ecologic transition model (Bricas *et al*, 2013). Moreover, by allowing to reduce the level of herd feed expenses, they could be a solution for farmers who are suffering from the variability of the agricultural product prices.

Conclusions

The study shows that a local social dynamics that favour grass can at least limit its decrease in the total UAA in a low-grass territory. It highlights the fact that agriculture should not be considered only for its production purposes, and that a greater dialogue between the farming community and the territorial institutions allows the attainment of multifunctional goals. In the long term, promoting grasslands could contribute to upgrading the breeding professional model, as it is known that, especially when the territory is composed of a mix of dairy farms and grain farms, breeders tend to discredit their profession, even when they earn a decent living (Fillonneau, 2012).

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Implications for conservation management of hay meadows; cutting dates

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Abstract

The Action plan for hay meadows in Norway is an instrument used to manage hay meadows of high biological value. The mowing management is regulated by a specific date and we aimed to assess the suitability of this date. We compared proportions of mature plants of species associated with semi-natural grasslands at the defined mowing date in 2014 and 2015. Numbers of mature species differed between the two years, but even in the warmest year the percentage of mature plants was only 44%. In order to plan a more successful agri-environmental scheme, the cutting date should be more site-specific and based on knowledge of the former practices that originally created and maintained the biological values.

Keywords: semi-natural grasslands, biodiversity, plant species, agri-environmental schemes

Introduction

Traditionally managed hay meadows are semi-natural habitats of high cultural and biological value, providing ecosystem services such as pollination, fodder, open landscape, and genetic resources (Bullock *et al.*, 2011; Totland *et al.*, 2013; Vinge and Flø, 2015). These grasslands include high species-richness of both plants and insects, but are danger-listed due to land use changes (Norderhaug and Johansen, 2011). In Norway the Action plan for hay meadows (Norwegian Environment Agency, 2009) has therefore been implemented as an agri-environmental scheme to manage these values.

An important aspect of high biodiversity in hay meadows is a late cutting date (Humbert *et al.*, 2012). This allows seeds to mature and disperse within the meadow and is highlighted in the action plan (Norwegian Environment Agency, 2009). However, farmers argue that a late cutting date results in less nutritious fodder. The management is regulated by an agreement between the authorities and the managers. All agreements allow for only one mowing which is to take place at the end of the season. Several agreements are specified with a predefined cutting date. Cutting is not allowed before this date. This is a common regulation in agri-environmental schemes that aim to protect species-richness in hay meadows throughout Europe (Dahlström *et al.*, 2013). In this project we aim to assess the suitability of the predefined cutting date on the maintenance of hay meadows of high value.

Materials and methods

We selected 28 hay meadows in the county of Møre og Romsdal which are implemented in the Action plan for hay meadows. Some agreements included predefined cutting dates on 10 or 15 July. The study sites were surveyed the week before these dates in 2014 and 2015. The first year was warmer and drier during the weeks before mowing (mean July temperature: 18.3 °C_{2014} and 13.1 °C_{2015} and July precipitation: 75.4 mm_{2014} and 82.2 mm_{2015}; data extracted from eklima.no).

During the two surveys we mapped the phenological stage of 34 plant species classified as associated with semi-natural grasslands. The phenological stage of the species was defined based on the proportion of mature plants (plants with mature seeds) in 1 m^2 plots. In order to investigate whether the phenological stages differed between species, between years and between hay meadows, generalized mixed models were built, using the lme4 package in the R 3.1.1 software (R Core Team 2015), and compared using likelihood ratio Chi-square tests.

Results and discussion

The results showed high yearly variation in the proportion of mature plants (Figure 1A), with the highest percentage in the warmest and driest year (2014). 22 species had more mature plants in 2014 and only five species were more mature in 2015 (Figure 1B). In 2014, 15 species had a proportion of mature plants >0.75 at the defined cutting dates. This means that only 44% of the species that the action plan aims to manage, had potential for new seed recruitment that year. In 2015, the number of species with proportion of mature plants >0.75 were only five. This indicates that for most species the predefined cutting dates were too early for the development of mature seeds. In the historic agricultural landscape, a farmer had to manage several meadows (Eriksson *et al.*, 2015). The most species-rich meadows were also, at that time, the least intensively managed. They were in resource-poor areas and often far away from farm settlements. In the mowing sequence, the least productive and furthest-away hay meadows were the last to be mown. In Sweden it was found that the end of the mowing period was mid-July in the south and mid-August in the north (Eriksson *et al.*, 2015).

The phenological stage varied among the species (Figure 1C) and even if the proportion of mature plants in the meadows did not differ (P=0.87), the stage reached by each species varied a lot between the plots (as seen in Figure 1C). Therefore, defining one cutting date might not be a good strategy for conserving the biodiversity. It is argued that regulations that set the same cutting date each year have caused population decline in hay meadows and that the present strict schemes do not comply with the management practices that have caused and maintained the high species richness in the past (Dahlström

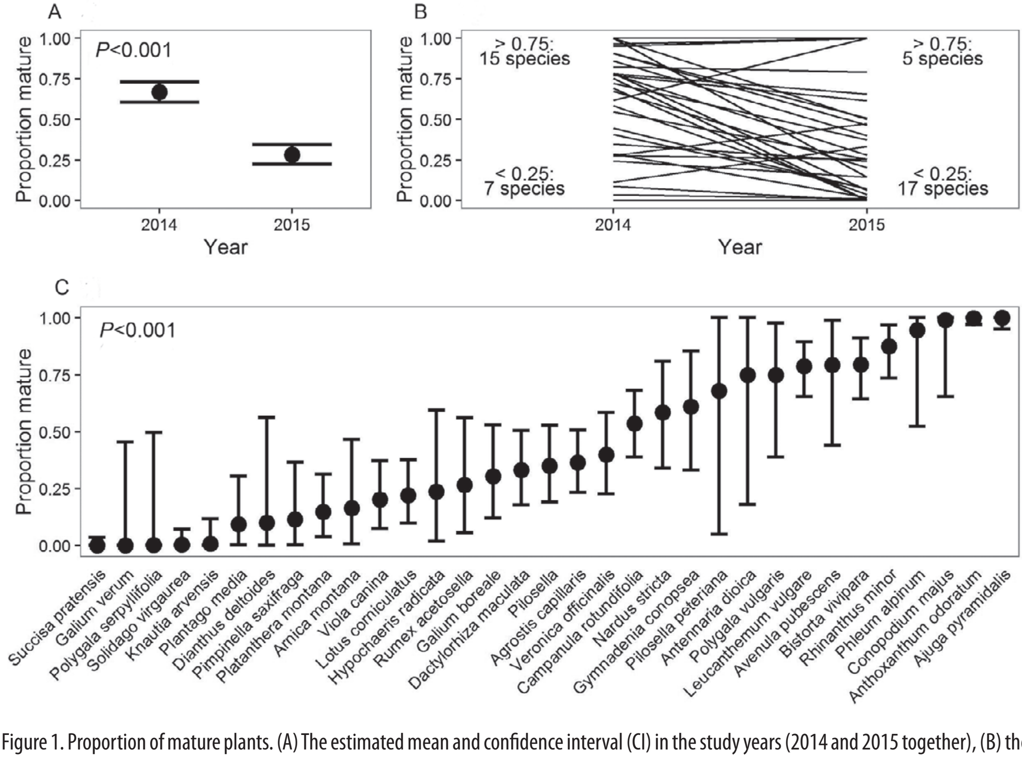

Figure 1. Proportion of mature plants. (A) The estimated mean and confidence interval (CI) in the study years (2014 and 2015 together), (B) the difference between the mean values of each species in the two years, and (C) the estimated mean and CI for each species.

et al., 2013). Historically, mowing dates varied between years (Eriksson *et al.*, 2015). Climate influences phenology, as our results show, and land use is and has been adapted to the weather. Swedish data show that historical mowing dates correlate with the start of flowering (Eriksson *et al.*, 2015). Our study includes a higher number of species associated with semi-natural grasslands compared to the Swedish study, and we found that the flowering period of these plants was not always the same. To allow seed production of the later flowering species, mowing has to take place later, even if not necessarily in all years. Mowing mid-July in some years, as suggested today, might help seed production of the few early-flowering species, as well as seedling and juvenile survival of the other species.

There is a need for an adaptive management of hay meadows. Regulations should not be static, they have to be adapted to the conditions of each meadow and based on knowledge of the former management practices that originally created and maintained the biological values that the Action plan for hay meadows aims to preserve.

Conclusions

Climate affects phenology and, in the semi-natural grasslands, different species mature at different times. Therefore, in order to maintain biological values in hay meadows, the management has to be adapted to each meadow and to the prevailing climate each year.

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The North Wyke Farm Platform: Data Portal

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Abstract

The North Wyke Farm Platform was established in 2010 as a UK national capability for collaborative research, training and knowledge exchange in agro-environmental sciences looking at agricultural productivity and ecosystem responses to different management practices for beef and sheep production in lowland grasslands. The Farm Platform Data Portal http://www.rothamsted.ac.uk/farmplatform provides the ability to preview and download data for specified time periods and has been populated with time series, livestock, field event and field survey data. A systems-based experiment on permanent pasture was implemented on three 21 ha farmlets in order to obtain baseline data in hydrology, nutrient cycling and productivity for 2 years. Since 2013 two farmlets were progressively modified in a 3-year programme through either (1) planned reseeding with grasses which have been bred for high water-soluble carbohydrate or deep rooting traits; or (2) sowing grass and clover-legume mixtures to reduce nitrogen fertiliser inputs. Each farmlet currently supports 30 weaned beef cattle and 75 ewes and their lambs that graze 5 hydrologically-isolated catchments instrumented to measure rainfall, soil moisture/temperature, water discharge and physical/chemical properties. Inputs of fertilisers, herbicides and farmyard manure are recorded and field surveys are conducted periodically.

Keywords: farm platform, database, data portal, livestock, grassland, field inputs

Introduction

The North Wyke Farm Platform (NWFP) is a farm-scale experiment with the aim of addressing agricultural productivity and ecosystem responses to different management practices for beef cattle and sheep grazing lowland grassland. The 64 ha site captures the spatial and/or temporal data necessary to develop a better understanding of the dynamic processes and underlying mechanisms that can be used to model how agricultural grassland systems respond to different management inputs. Here we describe the portal by which the data, collected since 1 April 2011, may be accessed.

Materials and methods

The underlying principle of the NWFP is to manage the three farmlets (Orr *et al.*, 2011) using (1) permanent pastures, (2) reseeding with long-term perennial ryegrass and white clover mixtures and (3) planned and regular re-seeding with innovative grasses. The connectivity between the timing and intensity of the different management operations, together with the transport of nutrients and potential pollutants from the NWFP is evaluated using various data collection exercises. The primary data collection strategy involves the use of a ground-based, wireless sensor network, where water flow, turbidity and chemistry are measured at 15 flume laboratories (5 per farmlet) that capture the water via a system of French drains installed at the edges of the catchments as described by Griffith *et al.* (2013). This sensor network also captures precipitation and soil moisture data at the centroid of each catchment and meteorological data (other than precipitation) at a single site. Greenhouse gas data (N_2O and CO_2) is also collected in three of the fifteen catchments (Cardenas *et al.*, 2016) using an automatic chamber system. Such high temporal resolution data sets (but with limited spatial resolution) are coupled with a secondary data collection strategy, for high spatial resolution data sets (but with limited temporal resolution). These latter data sets include field studies that provide information on soils nutrients and biodiversity.

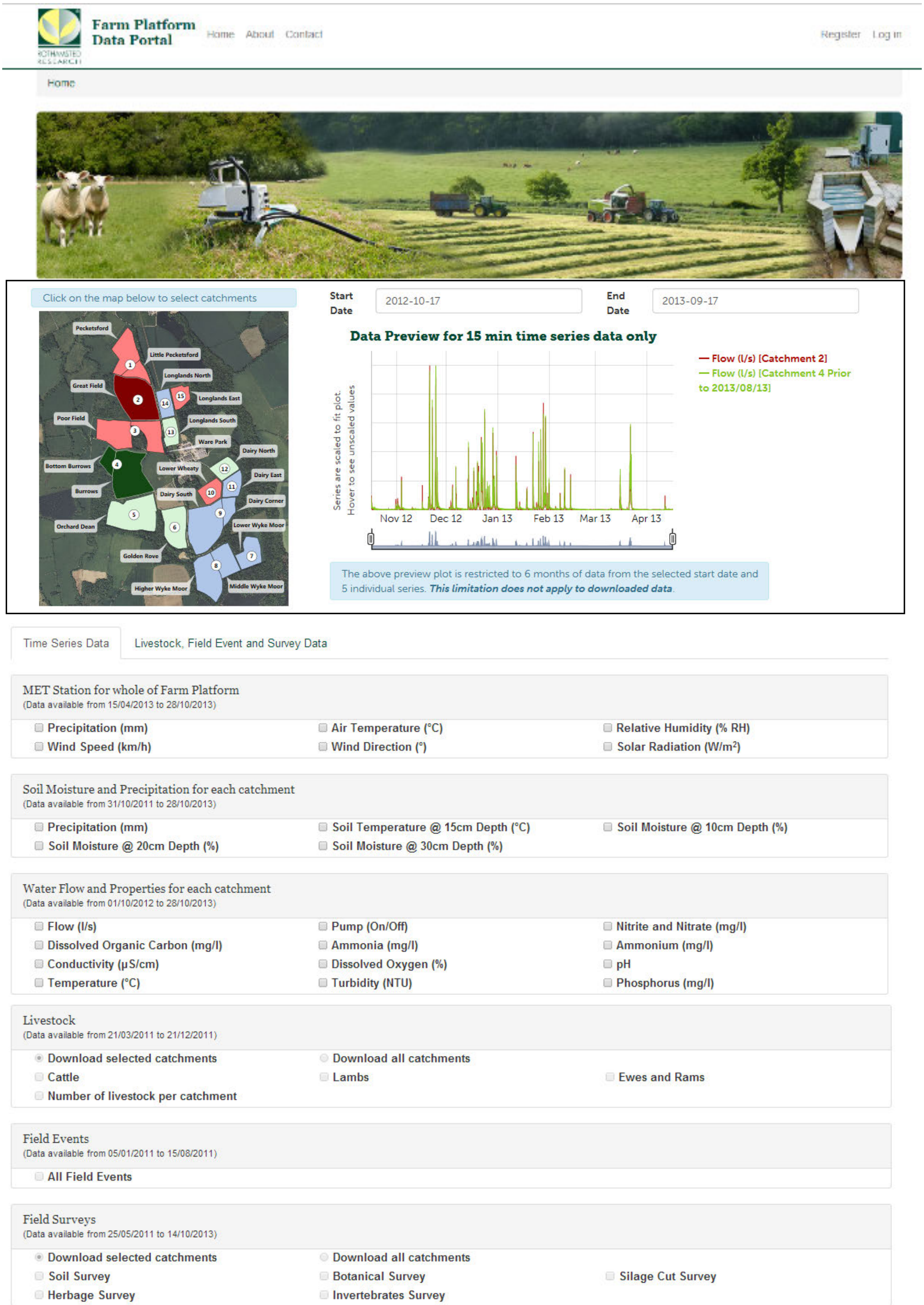

Figure 1. The North Wyke Farm Platform data portal.

Results and discussion

The data portal (Figure 1) may be freely accessed from the NWFP website http://www.rothamsted.ac.uk/farmplatform and provides registered users with the capability of selecting individual or groups of catchments and then exploring graphically the temporal changes in the Time Series Data (e.g. rainfall, soil temperature, soil moisture, water discharge hydrographs and chemical and physical attributes). Also, Livestock Data is available which comprises counts of the numbers of cattle, ewes and lambs located on each catchment each day, along with the location of individual animals. Livestock data also includes live weights and body condition scores, which are measured periodically, along with sales data (e.g. date of sale, carcase weights, conformation and fat class score, sales receipts) – all of which can be downloaded as .csv files. Field Events Data comprise dates and application rates of field inputs (e.g. fertiliser, farm yard manure, lime, herbicides) and field operations (e.g. mowing, silage making, ploughing, cultivations, seeding, rolling) which can be downloaded. Field Surveys provide information at geo-referenced locations on silage yield estimates made using a Haldrup (www.haldrup.de) plot harvester, along with co-located sampling-grid-based data on soil attributes (e.g. C, N, pH, organic matter), botanical species composition and invertebrates (e.g. tipulidae). The Data Portal also provides User Guides and Case Studies and links to useful external data sources.

Conclusions

The NWFP Data Portal provides a robust and powerful tool to visualise and obtain data from a highly-controlled lowland grassland beef and sheep system where the impact of the livestock on nutrients leaving the systems in discharge water and emissions are precisely quantified in a world-class facility. As a key member of the Global Farm Platform (http://www.globalfarmplatform.org/) the NWFP is contributing to a vision of sustainable and responsible production of ruminant livestock from pasture and forage-based systems.

Acknowledgements

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Evaluating ecosystem services in the life cycle assessment of grassland-based dairy systems

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Abstract

The integration of ecosystem services (ESS) in life cycle assessment (LCA) poses two main challenges: (1) how to integrate ESS within LCA, and (2) how to quantify the delivery of ESS. Several approaches have been proposed to integrate ESS in LCA: using multiple functional units, allocation to ESS, system expansion and introducing additional indicators. Some ESS are already directly or indirectly covered in LCA impact categories, while others remain to be added. The methods SALCA-biodiversity, SALCA-soil quality and the newly developed SALCA-landscape aesthetics will be applied to assess the environmental impacts and ESS from three grassland-based dairy production systems in Switzerland.

Keywords: life cycle assessment, ecosystem services, biodiversity, soil quality, landscape aesthetics

Introduction

Grassland-based farming systems provide multiple ecosystem services (ESS). The Millennium Ecosystem Assessment (2005) report defined ESS as the 'benefits people obtain from ecosystems' and distinguished provisioning, regulating, cultural and supporting services. Later CICES (2016) proposed a classification into (1) provisioning services, (2) regulation and maintenance as well as (3) cultural services. The delivery of high-quality feed for animals and subsequently animal products belongs to the most important provisioning services of grassland. The regulation and maintenance services of grassland are manifold, e.g. through biodiversity (habitat, species and genetic diversity), functional biodiversity and pest regulation, soil quality and fertility, symbiotic nitrogen fixation, prevention of soil erosion, of nutrient leaching and run-off, feeding sources for pollinators, recycling of animal manure, and the potential for carbon sequestration in the soil. Among the cultural services, the aesthetic value of the landscape is of primary interest. Häyhä and Franzese (2014) describe the challenges of ecosystem service assessment and conclude that using one single approach – like e.g. economic valuation – is not capable to capture all relevant aspect of ESS. Instead, they advocate approaches combining different perspectives and metrics. Combining ecosystem service assessment with life cycle assessment (LCA) is such an approach. However, this approach poses two main challenges which are discussed in this paper: (1) how to integrate ESS within LCA, and (2) how to quantify the delivery of ESS.

Integrating ecosystem service assessment within life cycle assessment

LCA provides a framework for the analysis of impacts on the environment. It can cover some ESS directly and indirectly, but for many ESS additional methods and indicators are needed (Zhang *et al.*, 2010). Koellner *et al.* (2013) proposed an 'ecosystem services depletion potential' including the biotic production potential, climate regulation potential, freshwater regulation potential, erosion regulation potential, and water purification potential. Since the systems provide multiple functions and services, they need to be considered as multifunctional. Several options are available in LCA to address the multifunctionality of agricultural systems:

1. Using multiple functional units to account for the various functions of the agricultural system (Nemecek *et al.*, 2011): area*time, agricultural products, income related indicator.
2. Using allocation, by dividing the environmental impacts between the products and ESS. For instance, Ripoll-Bosch *et al.* (2013) have analysed the greenhouse gas (GHG) emissions from different lamb production systems in the Mediterranean region. Extensive production systems tend to have high GHG emissions per kg of meat. Economic allocation was used to allocate the environmental impacts between meat production and the provision of ESS, thus considering the delivery of ESS as a co-product. The economic value of ESS was estimated by the agri-environmental payments to the farmers, considered as a proxy for the willingness of the society to pay for the ESS. Without considering ESS, the intensive system performed best; after the allocation the extensive system had lower impacts. Kiefer *et al.* (2015) used economic allocation including ESS to allocate environmental impacts between milk, meat and the provision of ESS. The value of the latter was hereby estimated by the subsidies paid to the farmers for the provision of ESS. Allocating part of the emissions to ESS reduced the environmental impact per kg of milk.
3. Using system expansion, where an alternative provision of ESS is subtracted (avoided impacts). An example would be mowing grass on Alpine pastures instead of grazing by animals, in order to maintain a grass cover.
4. Including ESS as additional indicators.

Using an area-related functional unit (option 1 above) can reflect some aspects of ESS delivery; however, the area used by agricultural production is only a poor indicator, not allowing a differentiation of various levels of ESS, and therefore this solution is not satisfactory. Allocating a part of the environmental impacts to ESS (option 2) requires an economic valuation of the latter, which can be approximated by subsidies or estimated by other economic valuation methods (e.g. Häyhä and Franzese, 2014). However, subsidies are defined in political processes and do not necessarily represent the true value of ESS. Furthermore, this approach means that the provision of ESS is associated with environmental impacts such as climate change or eutrophication. The system expansion approach (option 3) requires the definition of an alternative system for ESS provision, which is often difficult to determine and also debatable. Including ESS as additional indicators (option 4) allows for a detailed and differentiated assessment. This will be explained in the next section.

Quantifying ecosystem services within life cycle assessment

The environmental impacts of grassland-based production systems can be assessed by the LCA method, e.g. as implemented by the Swiss Agricultural Life Cycle Assessment SALCA (Nemecek *et al.*, 2010). Hereby, some ESS can be assessed within the LCA framework, either directly, like C sequestration, prevention of water pollution or prevention of soil erosion or indirectly, like symbiotic N fixation leading to less need for N fertilisers and to mitigation of related environmental impacts. For other aspects, additional indicators are needed.

Biodiversity is a basis for the provision of multiple ESS, and a higher diversity increases the chance to provide the required ESS, although there is not a simple relationship. The potential impacts of agricultural management on biodiversity are assessed by the SALCA-biodiversity method (Jeanneret *et al.*, 2014), using eleven indicator-species groups, namely flora of crops and grasslands, birds, mammals, amphibians, snails, spiders, carabids, butterflies, wild bees, and grasshoppers. The method distinguishes between several level of management intensity of grassland, as well as grazing and cutting grass.

Soil quality is assessed by the method SALCA-soil quality (Oberholzer *et al.*, 2012), which characterizes impacts of land management practices on the quality of arable soils by means of nine indicators covering soil physical, chemical and biological aspects: rooting depth of soil, macropore volume, aggregate stability, organic carbon content, heavy metal content, organic pollutants, earthworm biomass, microbial biomass and microbial activity. The model has been recently extended to better take into account the effects of grazing animals.

Landscape diversity and aesthetics will be assessed by the newly developed method SALCA-landscape (Roesch *et al.*, 2016). It is based on preference values for the aesthetical quality of different landscape elements. The diversity and seasonality of the landscape is considered (Schüpbach *et al.*, 2016).

Outlook

ESS provided by grassland systems can be quantified by biophysical indicators and integrated in the LCA approach. The challenges hereby are to assess the effect of the production systems on ESS on farm, to cover the multitude of ESS and also to include ESS of upstream stages, such as production of concentrate feeds. The presented method will be applied to analyse three dairy production systems, implemented in a trial is being carried out at the Hohenrain demonstration farm in Central Switzerland (Hofstetter *et al.*, 2014) and on 38 pilot farms in three regions of the Swiss lowlands.

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Pasture growth, structure and morphology during winter of diploid and tetraploid perennial ryegrass with or without white clover

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Abstract

An investigation into the growth and morphology of white clover (WC) and perennial ryegrass (PRG) overwinter was undertaken from November 2014 to February 2015. The experiment was a 2×2 factorial arrangement, consisting of two PRG ploidies and two WC contents, giving four treatments: tetraploid only, diploid only, tetraploid + clover, diploid + clover. Measurements included pasture dry matter (DM) yield (kg DM ha^{-1}), WC proportion, stolon mass, and tiller density. Pasture DM yield and WC proportion were measured every three weeks, the other measurements every six weeks. The average overwinter pasture DM yield and sward WC content did not significantly differ between ploidies. The inclusion of WC reduced pasture DM yield. Diploid swards (with or without WC) had a greater tiller density (+769 tillers m^{-2}) than tetraploid swards regardless of the presence or absence of WC. The implication of these observations is that inclusion of WC in a PRG sward can alter winter sward dynamics. The extent of this effect on spring performance remains to be determined, but based on the evidence from this study a superior performance from WC swards would not be expected.

Keywords: white clover, perennial rye grass, ploidy, overwintering, growth

Introduction

Grazed pasture is the cheapest source of feed for animal production (Finneran *et al.,* 2012), and the utilisation of increased quantities of grazed pasture at farm level can provide the basis of sustainable livestock systems. Perennial ryegrass (*Lolium perenne* L.; PRG) is the most common forage grass species used in Europe due to its regrowth capacity, grazing tolerance and high nutritive value for livestock systems (Wilkins and Humphreys, 2003). Perennial ryegrass ploidy has been the focus of recent research in terms of pasture production, nutritive value and animal production (Wims *et al.,* 2013). Renewed interest in utilising forage legumes exists due to their potential to increase pasture yield and nutritive value, the efficiency of conversion of pasture to animal protein and the substitution of nitrogen (N) fertiliser by biological nitrogen fixation to facilitate adaption to climate change (Lüscher *et al.*, 2014). Of these legumes, white clover (*Trifolium repens* L.; WC), is the most common and important species grown in temperate regions worldwide (Dewhurst *et al.*, 2009). Information on the overwinter survival of WC plants is limited, and so there is insufficient understanding of the underlying plant interactions that contribute to winter growth and persistence. Such knowledge could indicate key plant and management factors that enhance spring growth potential. Therefore, this study examined the growth patterns and structural characteristics during the winter period of diploid and tetraploid PRG swards with and without WC.

Materials and methods

The experiment was undertaken at Clonakilty Agricultural College, Cork, Ireland. The N application rate at the site was 250 kg N ha^{-1} per year. The experimental area was rotationally grazed at 2.75 cows ha^{-1} from February to November 2014 when all treatments were managed similarly. The swards used in this study form part of a larger farm systems experiment established in 2012, to examine the effect of PRG ploidy (tetraploid; diploid) with or without WC inclusion on spring milk production systems. The configuration was a 2×2 factorial design; two PRG ploidy treatments (tetraploid and diploid) and two clover treatments (with and without clover) resulting in four treatments (tetraploid only; (T), diploid only (D), tetraploid + clover (TC) and diploid + clover swards (DC)). In total, five blocks of four paddocks were used across the study site, amounting to 20 paddocks. Measurements included pasture dry matter (DM) yield, WC proportion, stolon mass, and tiller density. Pasture DM yield was estimated by cutting representative samples per paddock, and recording the length, weight, and heights of the cuts. Sward WC proportion was estimated by taking herbage samples and manually separating these into grass and clover components. Pasture DM yield and WC proportion were measured five times, every three weeks, and all other measurements three times, every six weeks, between November 2014 and February 2015. Data were analysed using Mixed Models in SAS (SAS, 2011). Terms included in the model were ploidy, clover content, measurement date and their interactions. Date was a repeated measure within paddock.

Results and discussion

The two grass ploidies did not differ significantly in average overwinter DM yield (Figure 1; T: 646 kg DM ha^{-1}, D: 678 kg DM ha^{-1}), or winter sward WC proportion (Figure 2; TC: 0.23, DC: 0.23). Pasture DM production was reduced by clover inclusion (P<0.001), with grass-only swards having greater average pasture DM yield (789 kg DM ha^{-1}) compared with grass-clover swards (534 kg DM ha^{-1}). Tiller density differed significantly between ploidies (P=0.007) as diploid swards had 773 additional tillers per m^2 than tetraploid swards (average: 4,195 vs 3,426 tillers m^{-2}) and also with clover inclusion (P<0.001) as grass-only swards had circa 1,953 more tillers m^{-2} than grass-clover treatments (4,789 tillers m^{-2} vs 2,837 tillers m^{-2}). Greater accumulation of stolon mass was indicative of a poorer tiller density; there was a significant negative relationship between stolon mass and tiller density across both grass ploidies. Previous studies mainly focus on growth during the main growing season, and so do not provide clear comparisons for the current study. However, the results from this study imply that PRG ploidy has no effect on the spread or success of WC plants in pasture during winter growth. This was despite the diploid swards generally having a greater tiller density than tetraploid swards and the inclusion of WC caused a reduction in tiller numbers due to competition for space. These observations are similar to the findings of Tozer *et al.* (2014) studying ploidy and Swift *et al.* (1993) studying WC inclusion effects.

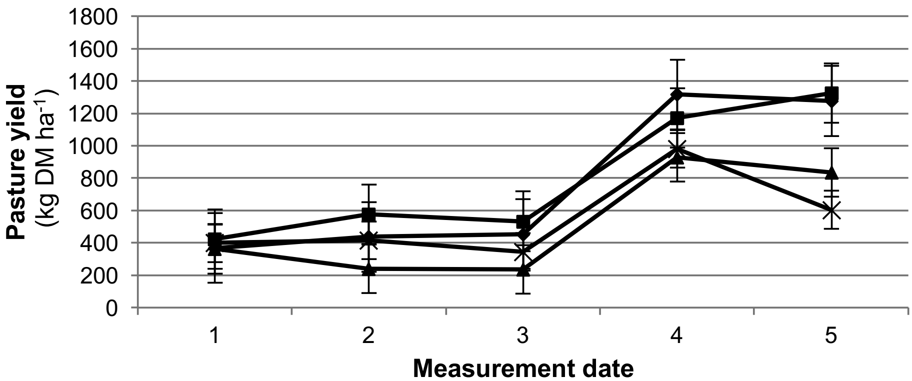

Figure 1. Average pasture yield (kg DM ha^{-1}) across all measurement dates for each treatment (tetraploid only (♦), diploid only (■), tetraploid plus clover (▲) and diploid plus clover (X), expressed by mean values; standard error represented by error bars).

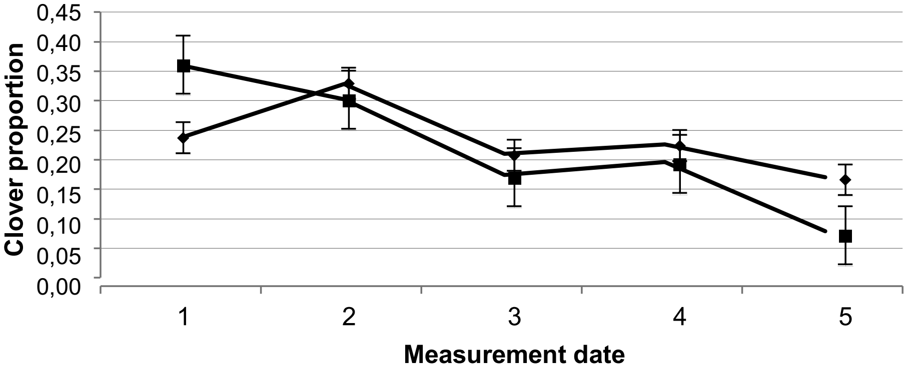

Figure 2. Average clover proportion across all measurement dates for each 'plus clover' treatment (tetraploid plus clover (♦) and diploid plus clover (■), expressed by mean values; standard error represented by error bars).

Conclusions

Perennial ryegrass ploidy had no effect on pasture DM yield or sward clover proportion during the winter, but had an effect on tiller density. However, white clover caused a significant decrease in pasture DM yield and tiller density during this period. The implications of these differences on spring pasture production remains to be determined.

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The 2014-20 CAP pillar 2 and conservation objectives – is there policy coherence for semi-natural grasslands?

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Abstract

Under the EU Habitats Directive, Member States (MS) are committed to maintaining Annex I habitats at favourable conservation status. Of the 231 habitat types, 58 are considered farmland habitats because they are dependent on or are associated with agricultural practices. CAP Pillar 1 payments are intended on all actively managed farmland, including permanent grassland. All of the 58 habitats referred to could be considered permanent grassland according to the CAP definition. Pillar 2 of the CAP includes the priority for restoring and preserving biodiversity, including in Natura 2000 areas and high nature value farming. It is reasonable to expect the CAP to be used in a manner consistent with the objectives of EU nature policy, and that Pillar 1 and Pillar 2 implementation will be designed to maintain livestock farming where this activity is needed for the conservation of Annex I habitats. This paper explores whether this is in fact the case. Focus is also placed on CAP context indicators that should monitor whether the conservations status of agricultural habitats is delivered.

Keywords: common agricultural policy, EU habitat directive, Natura2000, indicators

Introduction

Annex I of the Council Directive 92/43/EEC from 1992 on the conservation of natural habitats and of wild flora and fauna (also known as EU Habitats Directive) lists 231 habitat types of common interest. Together with sites according to the EU Birds Directive (Directive 79/409/EEC from 1979) both form the European Natura 2000 network. In article 3 (1) of the Habitats Directive, it is stated that this network shall enable the natural habitat types and the species' habitats concerned to be maintained or, where appropriate, restored to a favourable conservation status in their natural range. In this sense the conservation status of a natural habitat means the sum of the influences acting on a natural habitat and its typical species that may affect its long-term natural distribution, structure and functions, as well as the long-term survival of its typical species within a defined territory. Furthermore, a conservation status is taken as favourable when:

- its natural range and areas it covers within that range are stable or increasing; and
- the specific structure and functions which are necessary for its long-term maintenance exist and are likely to continue to exist for the foreseeable future; and
- the conservation status of its typical species is favourable.

Since no other adequate policy exist, and thus no other specific instruments to financially support habitats that depend on certain land use practises, the CAP is of crucial importance to achieve conservation aims as outlined in the EU Biodiversity strategy. This paper examines the following questions:

- Which is the significance of extensive grassland management practises to achieve legal conservation aims such as stated in the EU Habitats Directive?
- Are the CAP context indicators an appropriate instrument to measure or signal success or failure of the CAP as important instrument to implement Natura 2000 objectives?
- Does the new CAP pillar 2 contribute to the target of maintaining favourable conditions of grassland habitat types according to annex I of the EU Habitats Directive?

Materials and methods

The authors have followed public debate and outcome of the new forming of the CAP 2014-2020 on a European and as well on various national levels. Published data from EU and national sources have been analysed (EC, 2014, 2015). They also draw on expertise work that highlights the significance of extensive livestock systems for conservation and the demand for tailored but also practice-orientated support schemes within the new CAP (Keenleyside *et al.,* 2014, Luick *et al.,* 2015).

Results and discussion

Significance of extensive grazing to achieve European conservation aims

Annex I oft the EU Habitats Directive lists 231 habitat types of which 58 are considered farmland habitats because they are dependent on or are associated with agricultural practices (Halada *et al.,* 2011). Such habitats are almost exclusively extensive grasslands. This is even the case in Germany. With now prevailing modern, industrialized agriculture more traditional systems still using extensive grasslands are now often restricted to marginal and mountainous regions. There is much uncertainty whether present agriculture can assure the continuation of extensive systems. So far grazing systems are often not considered as being an appropriate instrument for conservation or there is controversial discussion. Although recently presented expertise showed that for 63 out of 92 Annex I habitat types in Germany (= 68%), large-scale extensive grazing could help to maintain favourable conservation targets of such habitats. To flag the priority: only 6.6% of the total grassland or 1.8% of the total utilised agricultural area in Germany can still be valued as grassland with conservation value for annex I habitats of the Habitats Directive (Jedicke and Metzner, 2012).

CAP context indicators

For the first time the common monitoring and evaluation framework (CMEF) of the EU will cover both CAP pillars (EC, 2014). Out of 45 indicators five are meant to evaluate and show the significance of biodiversity in agricultural systems: (1) sub-indicator of C.33 (farming intensity/areas under extensive grazing; livestock density <1 livestock unit ha^{-1} of forage area); (2) C.34 (Natura2000 area); (3) C.35 (farmland birds index); (4) C.36 (conservation status of agricultural habitats/grassland) and (5) C.37 (high nature value farming). Comparing provided data sets of indicators C.33 to C.37 (EC 2014) it can be concluded that there is a significant correlation between share of extensive grazing/grasslands and favourable conservation status of agricultural land (Figure 1). Extreme importance of extensive grazings for conservation correlates for Estonia, Italy, Latvia, Portugal and also for Sweden. In those countries, with more than 50% of grazing land characterized as extensive, up to 100% contributes to the favourable conservation status of agricultural land.

Signifiance of CAP pillar 2 for Natura 2000

For the new CAP period, 118 different rural development programmes (RDPs) have received acceptance by the EU (EC, 2015). Twenty Member States (MS) have installed one single national programme, eight MS have chosen to have more than one programme – in some cases to combine a main programme with a specific regional programme, e.g. for island regions, and in others to reflect federal administrative or/and geographical structures. Most important are agri-environment climate schemes with a must share of at least 30% of the total single RDP budget. However, the architecture of most measures shows that little to no attention is given to the specific demands of grassland habitats with conservation status. It is assumed that schemes have mainly been developed to comply with simplicity targets set by administration and controlling requirements, whilst ecological aspects have been suppressed.

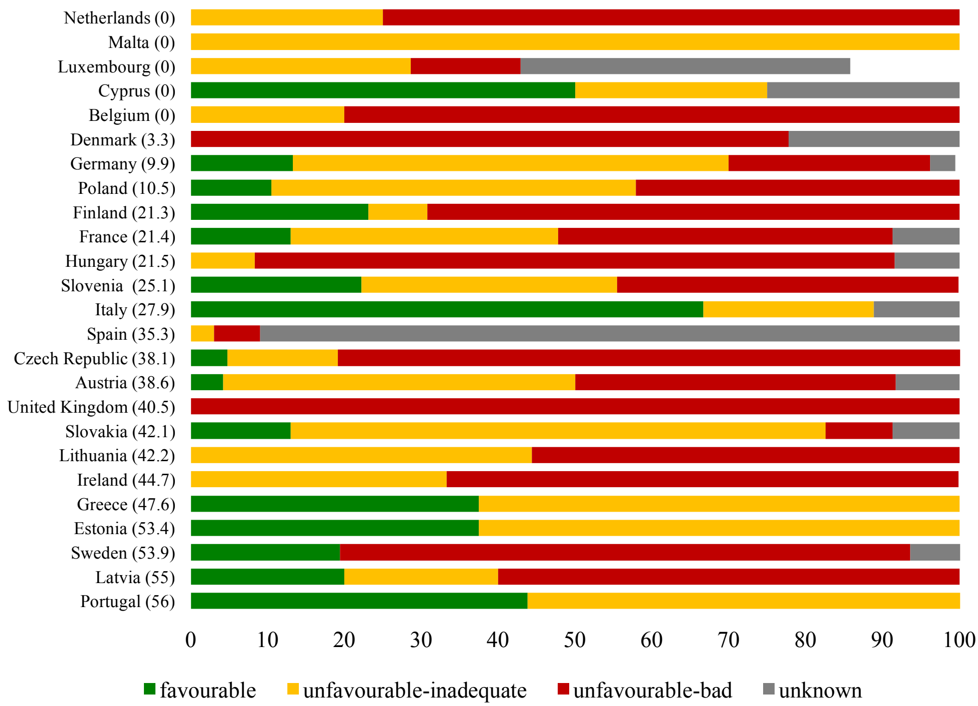

Figure 1. Data of CAP indicator C.36 – Conservation status of agricultural habitats (grassland) for the timeline 2000-2006. Countries are sorted from top to down according to their areas with extensive grazings as % (numbers in brackets) of total of utilised agricultural area (= CAP indicator C.33: Farming intensity, areas of extensive grazing; no data yet provided for Bulgaria, Croatia and Romania) (EC, 2014).

Conclusions

Despite clear data held by EC on the unfavourable condition of the majority of semi-natural grassland in Annex I of the Habitats Directive, another round of RDPs will in general fail to respond to the challenges of the EU Biodiversity Strategy, making very little use of the potentials of agri-environment-climate or Natura 2000 measures for improving the condition of these habitats. The new CAP Indicator on Farmland Habitats (grasslands) is weakened by lack of comprehensive monitoring of the condition of these habitats in many countries, but good monitoring systems are of no use if MS are not required to respond to what the indicator shows when designing or reviewing their RDPs.

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Influence of cutting date on botanical composition and yield of species-rich lowland meadows in south Germany

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Abstract

Species-rich lowland hay meadows are a characteristic landscape element of grassland in south Germany and are protected by the European Habitats Directive as 'habitat type 6510'. The conservation status of these meadows is frequently reported to be unfavourable – farmers are often unsatisfied with the biomass yield and forage quality. This study considers the effect of postponing the date of the first cut from mid-May to the beginning of July on plant diversity, forage yield and quality. This was investigated with four different cutting dates at two sites from 2013 to 2015. Annual mean dry matter yield (5.2 t ha^{-1}) was found to be only slightly affected by cutting date. The percentage of first growth in annual yield increased from 50% in the early cut to 63% in the late cut. Crude protein and net energy content declined by 43 and 21%, respectively. After 3 years, the number of species was highest in the mid-May cut, but only slightly lower in the later cuts. This suggests that early cutting may not be the only factor endangering biodiversity. However, it may take several years before these effects become evident in permanent grassland through suppressed propagation

Keywords: lowland hay meadows, cutting date, biodiversity, biomass yield, forage quality

Introduction

Species-rich lowland meadows have developed over the last decades as a result of a low-intensity grassland management. They belong to the *Arrhenatherion* alliances and are traditionally used as hay meadows. These species-rich meadows are usually found on moderately fertilised soils. Traditionally they are cut twice a year, the first cut with the onset of grass flowering. The European Habitats Directive 'Natura 2000' protects them as habitat type 6510 (European Commission, 2013), but despite this conservation status, a decline in area and botanical quality has been reported in recent years (Raufer *et al.*, 2015). Cutting date is considered a major factor in the maintenance of high grasslands diversity. Farmers tend to prefer earlier cuts to achieve high fodder quality. On the other hand, in some regions with surplus grassland, the intensity of management is too low (very late first cuts). Therefore, a field experiment was established to evaluate the influence of progressively staged first cutting date on vegetation, yield and biomass quality of habitat type 6510 grassland.

Materials and methods

In 2013, field experiments were established at two sites with habitat type 6510 grassland in a low mountainous area near Stuttgart, south Germany, one towards the top of the hills, the other in the foothills,10 km away. The higher site is referred to as 'Swabian Jura' (48.527N 9.532E, 774 m a.s.l., mean annual temperature 7.4 °C, mean annual precipitation 1,040 mm) and the lower as 'Foothills' (48.574N 9.442E, 470 m a.s.l., mean annual temperature 9.6 °C, mean annual precipitation 970 mm). The experiment was set up in a block design (plots 5×5 m, 3 replications). The date of the first cut was varied in four steps at each site (C1: mid-May, C2: end of May/beginning of June, C3: mid-June, C4: end of June/beginning of July). The date of the second cut was identical for all plots (mid-September). These cutting regimes were performed in the same way each year. The biomass of each cut was dried, ground and analysed for net energy (NEL), crude protein, fibre (acid detergent fibre (ADF), acid detergent

Table 1. Botanical composition and species number of first growth 2015.[1]

Site	Foothills				Swabian Jura			
Cutting date	**C1**	**C2**	**C3**	**C4**	**C1**	**C2**	**C3**	**C4**
Grasses %	58.1^{a}	64.2^{a}	62.0^{a}	63.1^{a}	58.5^{a}	72.2^{b}	68.7^{bc}	59.2^{ac}
Legumes %	4.1^{a}	3.3^{a}	3.3^{a}	2.6^{a}	4.6^{a}	4.1^{a}	1.8^{b}	1.9^{b}
Other herbs %	37.8^{a}	32.5^{a}	34.7^{a}	34.3^{a}	36.9^{ab}	23.7^{c}	29.5^{bc}	38.9^{a}
Species number	38.2^{a}	35.2^{ab}	35.7^{ab}	34.6^{b}	33.3^{a}	32.3^{a}	31.9^{a}	32.7^{a}

[1] Different superscript letters indicate significant differences between cutting dates at the same site ($P \leq 0.05$).

lignin (ADL)), K, P, Ca concentration (VDLUFA methods; Naumann *et al.,* 1997) and methane yield (Hohenheim biogas yield test). Prior to the first cut of each year, the botanical composition of the vegetation was estimated for each plot (dry matter yield percentage). Data analysis was performed using ANOVA and Fisher LSD test with the software R.

Results and discussion

The proportions of grasses, legumes and other herbs were not influenced by the cutting date at the site 'Foothills', but at 'Swabian Jura' the amount of herbs was reduced in C2 and C3 (Table 1). This was mainly an effect of a rapid decline in the annual herb *Rhinanthus alectorolophus* at this site due to inhibited seed propagation. The ensuing gap was filled by other herbs, such as *Galium mollugo,* in C1 and by grasses in C2 and C3. Where annual species were missing, no response of the vegetation to the variation in cutting date was seen at the end of the three-year period. The higher species number in C1 at the 'Foothills' site could be a result of lower shading of the herbs by tall grasses.

The biomass yield of all three years was only slightly affected by cutting date, with a trend towards highest yields in the mid-/end-of-June cuts. This is the time when hay making is traditionally carried out in these meadows. The earlier the cuts, the higher the proportion of the second cut in the yield (Figure 1).

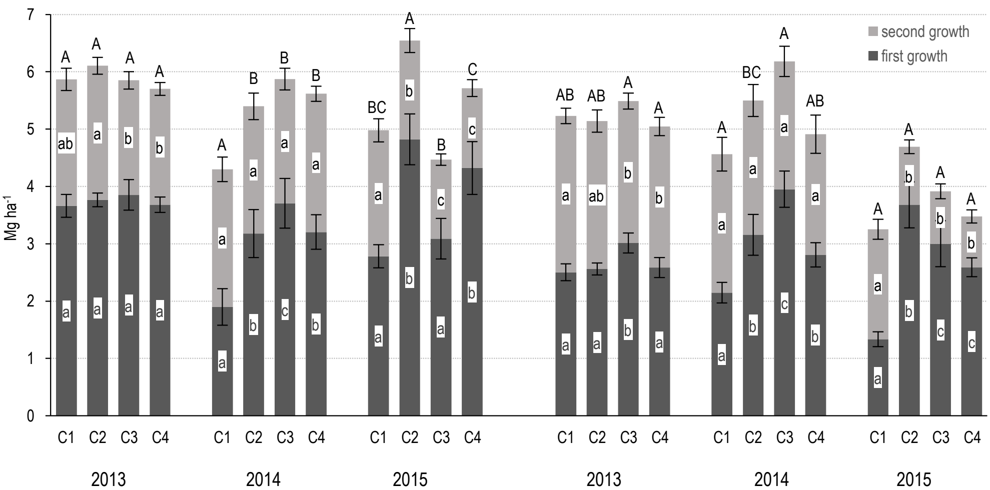

Figure 1. Dry matter yield of first and second growth in years 2013-2015 (significant differences between cutting dates at same site and year, first and second growth: abc, annual yield: ABC ($P \leq 0.05$).

Biomass quality is strongly affected by cutting date (Table 2). Net energy, crude protein, and P and K content of the first growth decline with later cutting date, whereas fibre (ADF, ADL) and Ca increase. There are considerable differences between the two sites due to the later start of the growing season at the higher altitude. Only C1 at 'Swabian Jura' meets the quality requirements of dairy cows; later cuts have limited use as animal feed. In contrast, the decline in suitability for biogas production (methane yield) was less pronounced from C1 to C2. Higher biomass yields in C2 result in the highest methane yields per hectare (Figure 1; Table 2). Biomass from late cuts is more suitable for combustion (Tonn *et al.,* 2010).

Table 2. Biomass (dry matter) characteristics of first growth 2015.[1,2]

Site	Foothills				Swabian Jura			
Cutting date	C1	C2	C3	C4	C1	C2	C3	C4
Net energy (MJ kg^{-1})	5.9^{a}	5.3^{b}	4.7^{c}	4.6^{c}	6.5^{a}	5.9^{b}	5.2^{c}	5.2^{c}
Crude protein (g kg^{-1})	117^{a}	83^{b}	72^{c}	67^{c}	150^{a}	101^{b}	93^{bc}	84^{b}
ADF	329^{a}	350^{b}	381^{c}	389^{c}	271^{a}	316^{b}	344^{c}	343^{c}
ADL	53^{a}	55^{ab}	58^{bc}	61^{c}	52^{a}	54^{ab}	57^{bc}	58^{c}
P	3.1^{a}	2.3^{b}	1.8^{c}	1.7^{c}	3.8^{a}	2.8^{b}	2.6^{bc}	2.4^{c}
K	24.0^{a}	18.6^{b}	16.0^{c}	16.0^{c}	28.8^{a}	22.2^{b}	19.6^{c}	19.3^{c}
Ca	8.0^{a}	8.1^{a}	8.7^{ab}	9.5^{b}	7.4^{a}	7.4^{ab}	8.6^{bc}	9.8^{c}
Methane yield Nm^3 kg^{-1} ODM	0.33^{a}	0.31^{b}	0.28^{c}	0.27^{d}	0.35^{a}	0.33^{b}	0.30^{c}	0.30^{c}
Methane yield Nm^3 ha^{-1}	915^{a}	1,475^{b}	873^{a}	1,181^{c}	464^{a}	1,208^{b}	918^{c}	769^{c}

[1] Different superscript letters indicate significant differences between cutting dates at same site ($P \leq 0.05$).

[2] ADF = acid detergent fibre; ADL = acid detergent lignin; ODM = organic dry matter.

Conclusions

The variation in cutting date was found to have only a slight effect on the vegetation of lowland meadows over a three-year period, but there was a considerable effect on biomass usability. However, a very different long-term effect of early harvest may be seen due to inhibited generative reproduction. Perennial grassland species need to be able to propagate to avoid aging of the population.

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Trade-offs between ecosystem services in managed and abandoned semi-natural grasslands

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Abstract

The effect of abandonment of sheep grazing management in semi-natural grasslands were studied in 14 sites in Norway. Data of species and vegetation composition, functional traits and pollination resources were used as indicators for nine selected ecosystem services (ES). The majority of the ES were negatively affected by abandonment of sheep grazing management. We therefore conclude that abandonment diminishes delivery of ES.

Keywords: grazing, supporting, regulating, provisioning, cultural, land use change

Introduction

Ecosystem services (ES) are the benefits humans obtain from nature (Millennium Ecosystem Assessment, 2005). Agricultural landscapes include mosaics of fields and grasslands ranging from monocultures to species rich semi-natural grasslands. Management of the semi-natural grasslands are relatively low intensive land use practices. The energy inputs are low but semi-natural grasslands provide multiple ecosystem services (ES); e.g. biomass, pollination, pest control, genetic resources, water quality, soil structure and cultural services such as recreation, aesthetics and tourism (Bullock *et al.*, 2011). The ongoing process of abandonment of extensive land use will influence provision of ES but little is known about the direction of these changes. The aim of this project is to assess potential effects of abandonment on ES in semi-natural grasslands in boreal ecosystems.

Materials and methods

Several studies have identified relations between vegetation characteristics and ecosystem services. Based on literature (De Bello *et al.*, 2010; Ford *et al.*, 2012; Pakeman, 2014; Vinge and Flø, 2015), we identified 13 ES indicators that can be calculated from terrestrial plant data (Table 1). We recorded all vascular plant species and vegetation structure in 112 subplots (4 m^2) in managed (sheep grazing) and abandoned semi-natural grasslands in 14 sites in West and Mid Norway. In addition, data on functional traits and plant species as allergy inducing pollen providers or resources for pollinators were extracted (LEDA; TRY; the Biological Records Centre's database of insects and their food plants; the Norwegian asthma and allergy association). Functional diversity was calculated using the R-package FD (Laliberte *et al*, 2014). The ES indicators were transformed into notations between zero and one using negative (abundance of allergy, cover of canopy and shrub layer, community weighted mean of leaf dry matter content) or positive linear transformation (the remaining indicators). The indicators are related to one or several ES and for each ES a bundle of indicators reflect the value of the ES. We therefore aggregated the indicators of each ES using weighted mean values of the notations. Then to assess the potential changes in ES due to abandonment, we compared the aggregated values of the ES in the managed and the abandoned semi-natural grasslands using linear mixed models (site as random). All analyses were done in the R software (R Core team, 2015).

Results and discussion

Our results show that grazing management generally has a positive effect on the deliverance of ES (Figure 1). The values of the supporting services nutrient cycling and genetic resources, the provisioning

Table 1. Ecosystem services (ES), indicators, and data. ES indicators: number of (NUMBER) plant species and flower colours, abundance of (ABUN) graminoids, legumes, herbs, allergy-inducing pollen producers (allergy), plant species attractive for *Hymenoptera* (Hymenopteran) and butterflies (butterfly), cover of (COVER) canopy and shrub layer, community weighted mean of (CWM) specific leaf area (SLA), leaf dry matter content (LDMC) and leaf nitrogen content (LNC) and functional richness of (FR) LDMC.

ES category	**ES**	**Indicator**	**Data**
Supporting	Nutrient turnover	ABUN legumes	Species composition
		CWM LDMC	Functional diversity
		CWM SLA	Functional diversity
	Allergy control	ABUN allergy	Species composition
	Genetic resources	NUMBER species	Species composition
Regulating	Pollination	ABUN butterfly	Species composition
		ABUN Hymenopteran	Species composition
Provisioning	Forage quantity	CWM LDMC	Functional diversity
		CWM SLA	Functional diversity
	Forage quality	CWM LNC	Functional diversity
		ABUN graminoids	Species composition
	Forage stability	FR LDMC	Functional diversity
Cultural	Aesthetics	ABUN herbs	Species composition
		NUMBER flower colours	Functional diversity
	Attractiveness	COVER canopy layer	Vegetation structure
		COVER shrub layer	Vegetation structure

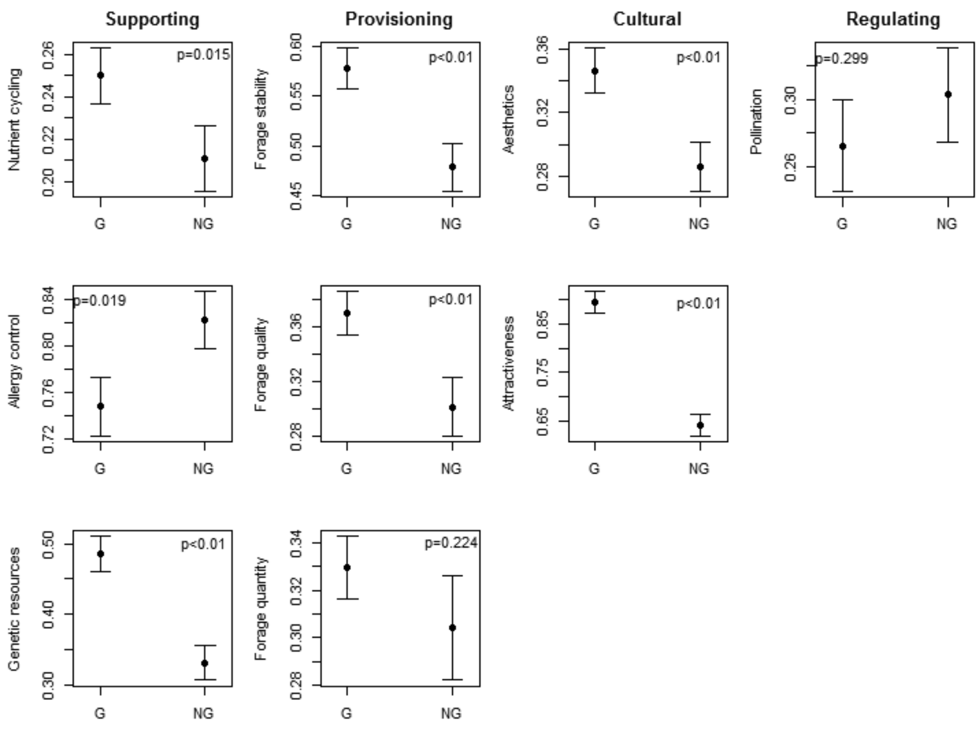

Figure 1. Mean and standard error of ecosystem services (supporting, provisioning, cultural and regulating) in grazed (G) and abandoned/not grazed (NG) semi-natural grasslands. *P*-values are based on likelihood ratio chi-square tests of mixed linear models.

services forage stability and quality, and the cultural services aesthetics and attractiveness were all higher in the managed semi-natural grasslands compared to the abandoned ones. Allergy control on the other side was highest in the abandoned grasslands. Different management regimes favour different ES (Ford *et al.,* 2012) and trade-offs need to be taken into account in conservation practices. Abandonment of grazing management in semi-natural grasslands results in encroachment and succession toward forest (Wehn *et al.,* 2011). Semi-natural grassland is a threatened nature type and its biological value is decreasing (Norderhaug and Johansen 2010), but this study show that semi-natural grasslands should be conserved also for supporting, provisioning and cultural ecosystem services.

Conclusions

Abandonment of management in semi-natural grasslands reduces the delivery of multiple ES.

Acknowledgements

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The effect of selected soil and climate parameters on multiple ecosystem services from abandoned and managed semi-natural grasslands

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Abstract

Climate, available resources and disturbance by agricultural land use influence ecosystem service (ES) delivery. In our project we studied how ES provision from managed and abandoned semi-natural grasslands vary along soil and climatic gradients. Information on climate (temperature and precipitation) and soil (pH-value and phosphorous content) were used to test whether ES varied along these environmental gradients. 13 ES indicators were calculated and assigned to nine ES. Some of the ES varied along the gradients, but the results indicate that the effects of soil and climate on ES are modified by agricultural land use.

Keywords: grazing, indicator, boreal ecosystem, land use change, plant species

Introduction

Semi-natural grasslands with high biodiversity provide several ecosystem services (ES; Bullock *et al.,* 2011). These ES are related to the biological characteristics of ecosystems and plant community surveys can therefore be used to measure ES at local scales (Kremen, 2005, De Bello *et al.,* 2010). Characteristics of plant communities can be species composition, functional traits, and vegetation structure. Delivery of one ES often depends upon several characteristics of the ecosystem, and within one ecosystem, bundles of ES interact (Lavorel *et al.,* 2010). Climate, available resources and disturbance by agricultural land use are some of the underlying environmental gradients that influence the characteristics of the communities in an ecosystem (Diaz *et al.,* 2007; Lavorel and Garnier, 2002). Therefore, to achieve knowledge about mechanisms that control ES delivery, the characteristics along environmental gradients have to be studied (Dorrough *et al.,* 2006; Lavorel *et al.*, 2011). The aim of this project was therefore to evaluate how ES provision from managed and abandoned semi-natural grasslands varies along soil and climatic gradients.

Materials and methods

We selected nine ES and defined 13 associated vegetation characteristics (indicators) (De Bello *et al.,* 2010, Duru *et al.,* 2012; Ford *et al.*, 2012, Pakeman, 2014, Vinge and Flø, 2015). The indicators are based on botanical surveys, functional traits (databases LEDA (Kleyer *et al.*, 2008), TRY (Kattge *et al.,* 2011)), information about the plant species as resources for pollinators (the Biological Records Centre's database of insects and their food plants) and allergy inducing pollen providers (information provided by the Norwegian asthma and allergy association). Plant community surveys were conducted on 112 subplots (4 m^2) within 14 Norwegian boreal sites including one manged and one abandoned semi-natural grassland. The ES indicators were transformed into notations between zero and one using negative or positive linear transformation using the TATALE tool (site http://umr-selmet.cirad.fr/en/products-and-services/proposed-products/tatale). Bundles of indicators related to one ES (see Table 1) were then aggregated using weighted mean values of the notations. The environmental data used in our analyses originate from soil samples (pH-value, available phosphorous (P)) and from the WorldClim database (http://www.worldclim.org/bioclim; mean annual temperature (MT), annual precipitation (AP)). Mixed linear modelling (site as a random) using the lmer4 package in the R software (R Core team 2015) was used to test the influence of environmental variables on ES in managed and abandoned semi-natural grasslands, separately.

Results and discussion

There was a positive effect of soil pH-value on genetic resources in both managed and abandoned semi-natural grasslands and on forage stability and aesthetics in the abandoned semi-natural grasslands (Table 1). Available P in the soil showed a negative influence on genetic resources and positive influence on allergy control, but only in managed sites. These results are linked to the higher species richness found in calcareous (Vandvik and Birks, 2004) and phosphorous poor soils (Dorrough *et al.,* 2006). Higher plant species richness causes higher diversity of herbs and flower colours, which are appreciated (De Bello *et al.,* 2010; Ford *et al.,* 2012), but it also increases the resilience of forage provision (Pakeman *et al.,* 2014). Fewer species on the other hand, give less allergy-inducing pollen producers (higher allergy control). We found lower delivery of pollination service with increasing level of phosphorus in the abandoned site but less nutrient turnover and forage quantity. No ES except allergy control varied along the climatic environmental gradients. In the abandoned semi-natural grasslands, allergy control decreased with both increased precipitation and temperature.

Table 1. The estimated effects (+: positive ($P<0.05$); -: negative ($P<0.05$); ns: ($P>0.05$)) of soil and climate on ecosystem services (ES) delivery in managed and abandoned semi-natural grasslands.

ES indicator	ES	Effect of soil				Effect of climate			
		Managed		Abandoned		Managed		Abandoned	
		pH	P	pH	P	MT	AP	MT	AP
Number of species	Genetic resources	+	–	+	ns	ns	ns	ns	ns
Abundance of legumes	Nutrient turnover	ns	ns	ns	+	ns	ns	ns	ns
Leaf dry matter content[1]									
Specific leaf area[1]									
Abundance of allergy pollen producers	Allergy control	ns	+	ns	ns	ns	ns	–	–
Abundance of butterflies attractive plants	Pollination	ns	ns	ns	–	ns	ns	ns	ns
Abundance of Hymenopteran attractive plants									
Leaf dry matter content[1]	Forage quantity	ns	ns	ns	+	ns	ns	ns	ns
Specific leaf area[1]									
Leaf nitrogen content[1]	Forage quality	ns	ns	ns	ns	ns	ns	ns	ns
Abundance of graminoids									
Functional richness of leaf dry matter content	Forage stability	ns	ns	+	ns	ns	ns	ns	ns
Abundance of herbs	Aesthetics	ns	ns	+	ns	ns	ns	ns	ns
Number of flower colours									
Cover of canopy layer	Attractive-ness	ns	ns	ns	ns	ns	ns	ns	ns
Cover of shrub layer									

[1] Community weighted mean.

Conclusions

Overall, we found a stronger influence of the environmental gradients on ES in the abandoned site compared to the managed semi-natural grassland, indicating that agricultural land use practices such as sheep grazing modify effects of climate and available resources in soil.

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Effect of different N, P, K fertilisation on plant species composition and species richness in an alluvial meadow

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Abstract

A fertilisation experiment in Admont (Austria) was set up in an alluvial wet meadow in 1946. Since then, 23 fertiliser treatments with various dose of N (nitrogen 80 kg ha^{-1} year^{-1}), P (phosphorus 35 kg ha^{-1} year^{-1}), K (potassium 100 kg ha^{-1} year^{-1}) and a non-fertilised control (Co) have been applied. In 2015 a botanical survey was conducted in selected treatments: (1) Co, (2) P, (3) K, (4) N, (5) KN, (6) PN, (7) PK and (8) NPK fertilisation. The highest number of plant species was revealed in P treatment, the lowest in Co treatment. Four groups of treatments with similar plant species composition were recognised on an ordination diagram based on RDA analysis: Co, P, K, and PK treatments form the first group, PN and KN treatments make up the second group, N treatment comprises the third group and NPK treatment constitutes the fourth group. Legumes *Trifolium repens* and *Trifolium pratense* had the highest abundance in the first group, *Agrostis capillaris* and *Plantago lanceolata* were supported by N, KN and PN treatments, *Trisetum flavescens* was positively affected by NPK fertilisation. N fertilisation supported short growing species, especially short grasses, P and K fertilisation supported legumes and other forbs.

Keywords: grassland, nutrients, cutting, biomass, diversity

Introduction

Alluvial meadows are grasslands that were formed in the flood plains of river valleys. As they were developed on mineral rich soils, their management was very intensive and in case of nutrient impoverishment they were fertilised (Hrevušová *et al.,* 2014). Applied management of alluvial meadows should provide not only sufficient forage as well as other natures of ecosystem services (e.g. species diversity). Long-term grassland experiments are an indispensable tool for understanding production and ecosystems services functions. The Admont Grassland Experiment (AGE) belongs to the oldest still-running grassland fertiliser experiments in the Central Europe. In 2015 we carried out a botanical survey in the AGE to specify in more details the effect of chosen applied nutrients and their combinations on the vegetation.

Materials and methods

The long-term field fertilisation experiment was established in 1946 in Admont, province of Styria (Austria) (47°34`52``N and 14°27`04``E, 635 m a.s.l.). The average annual precipitation at this site is 1,227 mm with a long-term mean annual air temperature of 6.8 °C. The parent material of the site is alluvium, the soil type is a gleyic, brown alluvial soil. The dominant species are *Agrostis capillaris*, *Anthoxanthum odoratum*, *Trisetum flavescens*, *Leontodon hispidus*, *Plantago lanceolata* and *Trifolium pratense*. According to Austria flora (Fischer *et al.*, 2008) the species were a priory categorized to short and tall grasses, short and tall forbs. Species with a mean height ≥0.5 m were classified as tall whereas those below this threshold were classified as short.

In 2015 eight out of 23 different fertiliser treatments with various dose of N (nitrogen 80 kg ha^{-1} year^{-1}), P (phosphorus 35 kg ha^{-1} year^{-1}), K (potassium 100 kg ha^{-1} year^{-1}) and a non-fertilised control (Co) were selected for comprehensive botanical survey: (1) unfertilised control (Co), (2) P, (3) K, (4) N, (5) KN, (6) PN, (7) KP and (8) NPK fertilisation.

AGE was established in four permanent, randomized blocks, using rectangular plots with a size of 2.9×7.1 m each. All treatments are cut regularly three times a year (around 25 May, 20 June and 30 September, depending on the particular weather and growing conditions). In May 2015 just before the first cutting date, the cover of all vascular plant species was recorded using the percentage scale of the whole plots. Redundancy analysis (RDA) in the CANOCO 5.0 program (Ter Braak and Šmilauer, 2012) was used to evaluate multivariate vegetation data in the year 2014. The blocks were treated as covariables to restrict permutations into blocks. A mixed linear model (one way ANOVA) with block as random factor was used for evaluation of number of vascular plant species, cover of short and tall grasses, cover short and tall forbs.

Results and discussion

The mean number of all vascular plant species ranged from 37.8 (P) to 28.3 (Co) per plot (Table 1). However, in P treatment on average 19.5 species showed a cover less than 0.5%. The low number of vascular plant species in the control treatment was probably caused by long-term cutting management with remarkable biomass removal. A lot of nutrients were depleted with removing biomass during the previous 69 years with tree time defoliation a year, resulting in an unexpected decrease in number of plant species. Besides that in Co treatment a large mean cover of mosses (26.2%) occurred. The highest species richness revealed in P treatment is not in accordance with the results of Janssens *et al.* (1998) that associate high level of plant available P with low plant species richness. Probably one-sided fertilisation of phosphorus in P treatment in our experiment is responsible for low productivity of above ground biomass which is usually connected with a negative relationship with species richness.

The results of RDA from the year 2015 showed significant (P<0.001) effects of treatments for the first ordination axis and all ordination axes in plant species composition. The percentage of explained variability by the first axis and all ordination axes was 26.7 and 55.6 respectively. Four groups of treatments with similar plant species composition were recognised on an ordination diagram based on RDA analysis (Figure 1): (1) the unfertilised control, P, K, and PK treatments form the first group; (2) PN and KN treatments make up the second group; (3) N treatment comprises the third group and; (4) NPK treatment constitutes the fourth group. For example legumes *Trifolium repens* and *T. pratense* had the highest abundance in the first group, *A. capillaris* and *P. lanceolata* were supported by N, KN and PN treatments, tall grass *T. flavescens* was positively affected by NPK fertilisation.

As frequent defoliation generally supports short plant species because of lower competition and better light conditions, the fixed cutting frequency of three times a year was probably responsible for higher cover of short species (Table 1) in our experiment. The highest cover of short plant species was observed in N treatment (100.7%) together with the highest cover of short grasses (66.8%). On the other hand

Table 1. Mean proportions (%) of tall grasses, short grasses, tall forbs, short forbs and mosses and mean number of all plant species in the studied treatments. Numbers represent average of four replicates ± standard error of the mean; P = probability value.

Treatment	Tall grasses	Short grasses	Tall forbs	Short forbs	Mosses	Number of all plant species
Co	4.3±1.6	25.0±5.9	21.0±4.7	33.0±10.4	26.2±6.8	28.3±1.4
P	24.8±6.5	21.0±1.1	34.3±4.8	33.4±3.4	–	37.8±1.1
K	2.1±0.6	24.9±3.1	29.0±2.4	45.1±5.6	4.8±4.2	29.5±1.3
N	4.6±1.0	66.8±3.2	8.8±1.8	33.9±6.0	–	31.8±2.3
KN	6.8±0.9	43.3±7.9	20.9±2.7	44.7±10.1	–	33.3±0.9
PN	24.4±2.9	35.7±3.5	12.2±2.4	45.5±7.0	–	33.3±1.1
PK	16.4±2.8	23.7±4.3	41.9±0.6	34.9±1.6	–	35.0±1.5
NPK	31.7±4.7	14.7±2.8	24.8±3.9	52.7±7.3	–	33.3±1.2
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	0.003

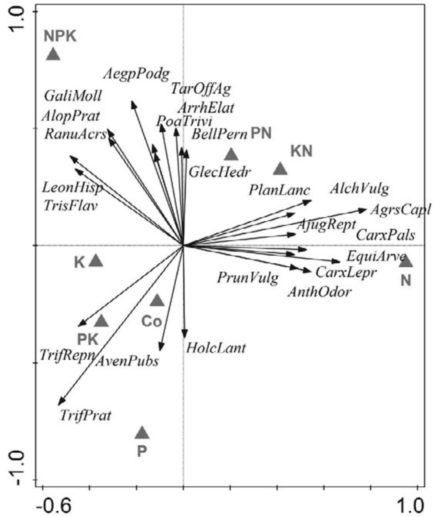

Figure 1. Ordination diagram showing the results of a redundancy analysis of vegetation data. For treatment abbreviations see materials and methods section. Species abbreviations are based on first four letters from genus and four letters of species name.

tall species were mostly supported by P treatment (59.1%) as well as by the other treatments with P application – PK treatment (58.3%) and NPK (56.5%). In all these treatments the cover of grasses was lower than the cover of forbs. The lowest covers of grasses were recorded in K (27.0%) and in the control (29.3%) treatments. Support of legumes cover by P (30.1%), K (27.8%) and PK (40.2%) application in our experiment is in agreement with results of Hrevušová *et al.* (2014). As N fertilisation supported grasses and P, K fertilisation supported forbs the combined application of N, P, K fertilisation showed intermediate influence on plant species composition.

Conclusions

The results from AGE showed that both plant species richness and plant species composition were affected by applied fertilisation. The highest number of plant species occurred in the P treatment, whereas the lowest came up in the unfertilised control. Long-term N fertilisation supported short plant species, especially short grasses, whilst P and K fertilisation supported legumes and other forbs.

Acknowledgements

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Grazer effects on plant species richness and tree debarking within orchard pastures

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Abstract

Orchards are traditional agroforestry elements in agricultural landscapes and valuable for nature conservation in Europe. However, their diversity is endangered due to farmland intensification or abandonment. Grazing management promotes plant species diversity but grazers are suspected to harm trees through debarking. We studied 42 orchards within the Rhenish uplands (Germany) and compared horses, cattle and sheep grazing. We analysed how grassland diversity is promoted by the presence of trees and modified by grazing management, as well as how grazers may impact trees. Plant species richness benefited from tree presence, but was not affected by tree-cover and only slightly by grazer species, whereas grazing intensity showed significant negative effects. All grazer species significantly increased recent debarking in the absence of individual tree-protection. Therefore, maintaining species diversity and long-term tree persistence in orchards does not primarily depend upon grazer species, but more especially on grazing intensity and tree-protection.

Keywords: orchards, agroforestry, grazer species, debarking

Introduction

Orchards are traditional agroforestry elements in agricultural landscapes, combining grassland and planted fruit trees. They are of great importance for biodiversity conservation, but endangered due to farmland intensification or abandonment (Garbarino and Bergmeier, 2014). Grazing evolved as an important management strategy over the last decades. However, research on grazing and debarking in German orchards is lacking. Since orchards have usually not experienced grassland intensification practices, they have relatively high plant species richness (SR). Furthermore, the presence of scattered trees in grasslands is known to promote niches with considerable variation in light availability and soil properties which enables species coexistence (Garbarino and Bergmeier, 2014). Therefore, in this study, we tested the hypotheses that (1) plant SR in grassland is increased by tree presence and depends on tree-cover. Grazers affect plant SR due to specific selectivity, nutrient deposition and trampling, which is known to differ between grazers and to depend on grazing intensity (Rigueiro-Rodríguez *et al.,* 2009; Wrage *et al.*, 2011). Hence, we hypothesized that (2) plant SR differs between grazer species and is affected by grazing intensity. As grazers are suspected to harm trees due to browsing and debarking (Plieninger *et al.*, 2015), we tested the hypotheses that (3) grazers species differ in the amount of recent debarking depending on grazing intensity and tree-cover, and that (4) tree-protection helps to avoid debarking.

Materials and methods

In 2014, we studied managed orchards of 40 farmers within the Rhenish uplands (Germany). 42 paddocks were arranged in 14 triplets, while each triplet comprised three neighbouring paddocks of similar site conditions (soil, slope, altitude) grazed by cattle, horse and sheep, respectively. To ensure vegetation equilibrium, only paddocks were chosen that experienced the same management since at least 5 years.

At all sites a subplot of 1,250 m^2 with at least 3 trees was randomly established. In addition, two transects (2×35 m) were chosen for vegetation analysis, one diagonal through the subplot, including all tree-introduced microhabitats (shady areas under crowns, ecotones) and the other located on the same paddock but not influenced by trees at all. All grassland plant species in transects were identified. SR per paddock was defined as cumulative number of species of both transects. Further, a regional set of species indicating high-nature-value (HNV) (BfN, 2016) was used to obtain information on biodiversity value of the study sites.

Recent debarking (less than 1 year) was estimated for all trees within each subplot as a percentage of the trunk. Protectors were recorded as presence/absence data. Tree-cover was estimated by digitizing tree crowns (in m^2) on an aerial picture (geobasis.nrw, 2012) using ArcGIS10.3 and related to the ground area of the subplot. Information on site management was obtained by interviewing farmers. Grazing intensity was estimated as live-weight unit grazing days per hectare and year (LUGD ha^{-1} a^{-1}, with one livestock unit = 500 kg). Statistics were performed in R 3.2.2 applying mixed effects models, lme in nlme package for vegetation data and lmer in lme4 package for recent debarking data. In all models we used livestock, LUGD ha^{-1} a^{-1}, and tree cover as fixed effects. Transect area and protector presence, were used as additional fixed effects for vegetation and recent debarking analysis respectively. In all models site was nested in triplet as random term. Recent debarking was fitted to binomial error distribution with a logit link. Most important fixed effects were obtained using the model averaging approach (MuMIn Package) best models were selected by AICc by delta <2. Residuals were visually checked for heterogeneity in final models.

Results and discussion

On 42 orchards, we observed a total of 145 grassland species. Tree-cover varied between 10 and 60%, with a mean of 23% and a significantly higher cover on cattle pastures. Mean grazing intensity did not differ significantly between grazer species but showed remarkable variability especially on horse pastures with mean 572 LUGD ha^{-1} a^{-1} (±464 standard deviation). Directly comparing vegetation of transects in tree-areas and areas not influenced by trees on the same paddocks, models showed significantly higher SR in tree-areas (Figure 1B).

Trees introduce heterogeneous microhabitats which enable species with different requirements to sustain, which Schmiedgen *et al.* (2016) confirmed in the current proceedings using the same paddocks following a design focusing on microsites. However, neither SR nor richness of HNV-species were affected by tree-cover, so dense tree-population does not promote SR per se. We did not find significant grazer species

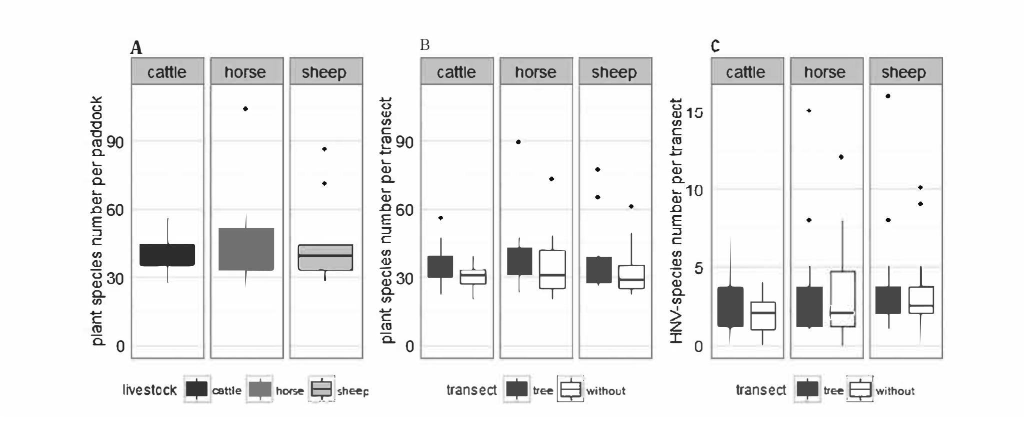

Figure 1. Variability of cumulative number of species per paddock (A) and species richness (B) as well as high nature value species richness (C) on transect scale (areas with trees or without trees), comparing grazer species. Boxplots present upper and lower quartiles around Median.

effects on SR at the transect scale or in cumulative SR per paddock. But there is a remarkable variability and slightly (P<0.07) higher SR in horse grazed paddocks (Figure 1A). However, grazing intensity did affect SR. Models showed a tendency (P<0.07) for a negative influence on SR on transect scale and a significant (P<0.01) negative effect on paddock scale. High stocking rates are known to decrease SR as livestock becomes less selective and tend to homogenize swards (Wrage *et al.*, 2011).

Recent debarking was not common in observed orchards, since only 8% of the analysed trees (n=457) showed it. We did not find differences among grazer species, all grazers significantly increased recent debarking in absence of individual tree-protection (Table 1). Therefore, in short-term grazers debarked a small proportion of tree-population, but the presence of protectors illustrates the importance to avoid future persistent stand damages.

Table 1. Summary of the Generalized Linear Mixed Models fitted for recent debarking as the response variable.

Response variable	Predictors	Importance	Factors	Coeff.	Standard error	z-value	P-value
Recent debarking (%)	intercept			-3.575	0.294	11.698	<0.001
	protector	1.00	yes	-1.149	0.532	2.072	0.038
	tree cover	0.20		-0.342	0.503	0.651	0.515
	livestock	0.18	horse	0.284	0.407	0.667	0.505
			sheep	-0.449	0.467	0.920	0.358
	Liveweigth unit	0.17		-1.2e-4	4.5e-4	0.259	0.796

Conclusions

In orchards, maintaining species richness and avoid debarking does not primarily depend upon grazer species, but rather on stocking rates and tree-protection. Therefore, in the absence of cattle or sheep, extensive grazing management with horses might help to maintain orchards. This might be reconsidered in strategies for biodiversity conservation.

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Persistence of legumes-based grasslands: some features for synergy between ecosystem services

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Abstract

Legume-based temporary grasslands, common in ruminant farms in France and other European areas, can ensure high-quality forage production and nitrogen self-sufficiency. Improving their persistence, i.e. maintaining their ability to produce high-quality forage beyond 4-5 years, requires managing equilibrium among forage species, which remains a challenge. This communication relates the initial results of a historical survey on vegetation dynamics in sown grasslands (Feader SOS-Protein-4AgeProd-SP3). Long-term grasslands can be found in a wide range of free-draining soil and oceanic climate conditions. Sward dynamics of these long term grasslands are presented and discussed in relation with their management.

Keywords: sown grassland persistence, multispecies, legumes, nitrogen, vegetation dynamics

Introduction

Sustainable livestock systems rely increasingly on 'on-farm' production of high-quality forage. Increasing the persistence of grass-legume grasslands beyond 4-5 years can ensure better nitrogen (N) self-sufficiency and help increase delivery of ecosystem services (Tilman *et al.,* 2006): biomass and high-quality forage provisioning, regulation of water and air quality by decreasing N losses and increasing soil N and C storage, lower use of N fertilizers and higher biodiversity (soil, plant, fauna). Equilibrium among legumes and other grassland species, a key issue for grassland persistence and N autonomy, is difficult to manage, since it depends on the ability of species to co-exist and complement each other in the long term. This ability depends on interspecific competition, which results from (1) the intrinsic longevity of species; (2) their ability to cope with soil and climatic conditions in interaction with management, e.g. with soil mineral N status (Schwinning and Parsons, 1996); and (3) species growth strategies in time (earliness of growth in the spring, summer growth strategy) and space (rooting depth, vertical vs horizontal shoot colonisation).

The participatory research program Feader SOS-Protein-4AgeProd-SP3 was set up to address two questions: (1) what main factors explain persistence of multispecies grasslands in lowland dairy systems in western France; and (2) what management can increase grassland persistence and favour development of a productive permanent grassland? The project includes two complementary work packages, combining 2 observatories (WP1) with experimental trials in farms (WP2) to identify factors critical for persistence and to characterize grassland ageing precisely. This paper presents initial results of the historical survey.

Materials and methods

The historical observatory included plots with sown grasslands more than 7 years old in 2015, managed by farmers who began recording their practices at sowing, while a new observatory will include 2-3 year old grasslands to be observed over 4 years. The data collected are (1) field characteristics: location, climatic zone, soil type and depth, initial species sown and current botanical composition (species and their cover in summer 2015 according to the method of de Vries *et al.* (1959); (2) questions about production and management, i.e. fertilisation, cutting and/or grazing periods, animal type and stocking density; and (3) a set of open qualitative information, e.g. what do you consider a good/bad grassland? Can you describe steps in grassland ageing? How do you assess production and quality?

Results and discussion

The results of the historical observatory include 35 grasslands in Brittany and Pays de la Loire considered to be in 'good state' by farmers on 28 farms. Their distribution covers a wide gradient of oceanic climate conditions (from mild to cold in winter, from wet to dry in summer). In our sample, mean grassland age was 12 (±5.5) years, with 50 and 37% being more than 10 and 15 years old, respectively, confirming the potential for old sown grasslands to keep a high productive potential.

For nearly all farmers 'good state' meant (1) good production: for most of them 6-8 tons dry matter (DM) ha^{-1} yr^{-1}, i.e. about 75-80% of young grassland production (calculated from animal needs); (2) 'good' balance between grasses and legumes, in which 'good' was defined differently (e.g. >30% legumes, 50-50, several species); and (3) a paddock grazable during a long period of the year (biomass, quality, soil with good bearing capacity). In contrast, a 'bad state' for most of them meant decreases in production, often attributed to a lack of legumes (as most plots were fertilised with mainly compost or cattle manure). All farmers mentioned the occurrence of bare soil patches and the development of weeds, such as *Rumex* sp., *Cirsium* sp., dicots (*Taraxacum* sp., *Plantago* sp., etc.) or grasses (*Agrostis stolonifera*, *Holcus lanatus*).

The mean number of sown species (cultivars not included) was 3.25 (±1.5), with 2 species in 30% of plots, 3-4 species in 30% of plots and ≥5 species in the remaining 40% of plots. The final botanical compositions (Table 1) showed a large increase in species diversity in all cases, with a mean increase over time of 5.6 (±4.4) to reach the mean number of 10 (±4). Sown species were always observed in the final botanical composition of the grasslands. The total number of species observed in 2015 was 39 (regional diversity), the maximum number in one plot being 22. The number of species in 2015 was not correlated with initial diversity of the plot (R^2=0.02 with a linear model) nor to grassland age (R^2=0.002).

Legumes were observed in all plots, with *Trifolium repens* occurring in all plots (representing 1-48% of vegetation) and *Trifolium pratense* in 20% of plots. However, their abundance in the vegetation exceeded 30% in only 25% of plots. Grasses remained highly dominant, the top five being the species initially sown: *Lolium perenne* (86% of plots, with a mean abundance in the vegetation of 23%, which is relatively low, as is commonly observed in Pays de la Loire during dry summers), *Festuca arundinacea* (63% of plots), *Dactylis glomerata* (51% of plots) and two grasses common in grazed grasslands: *A. stolonifera* (63% of plots) and *H. lanatus* (57% of plots). They were followed by *Poa trivialis, Agrostis tenuis* and *Poa pratensis* (>20% of plots). The most frequent and abundant unwanted species was *Agrostis stolonifera*, whose abundance exceeded 20% of vegetation in 20 plots). *Rumex* sp. and *Cirsium* sp. were rare (in less than 5% of plots and often scarce). The most frequent dicots were *Taraxacum* sp. (83% of plots), *Hypochoeris radicata* (40% of plots) and *Plantago lanceolata* (34% of plots, locally abundant). These botanical characteristics were consistent with the farmers' own assessments.

Table 1. Mean final botanical composition in the 35 grasslands (2015).

No. species	% plots	% legumes in plant cover	% plots	% dicots in cover	% plots
≤5	9%	<5%	3%	<5%	43
6-10	54%	5-15%	37%	5-10%	20
11-15	23%	16-30%	37%	11-20%	26
>15	14%	31-50%	23%	21-25%	11

Soils commonly had good structure and were free draining, while soil texture and depths varied greatly. At most sites, rooting systems were well developed in the 20 upper cm. No or moderate soil compaction (10 cm depth) was observed in more than 80% plots, while strong compaction was found (at 25-30 cm) in only 17% of the plots, preventing root development below. No correlation (linear model) was observed between soil depth and grassland age, final number of species, or abundance of legumes or dicots. Concerning management, most grasslands were fertilised only with manure once every 2-3 years (15% with mineral NPK), and rotationally grazed by dairy cows, with a mean of 5 (range = 3-11) grazing cycles per year. Two-thirds of plots were cut at least once every 2 years. This explained the low presence of erect species, such as alfalfa, Italian ryegrass (sown in 5% of plots) or red clover (sown in 12% of plots) less tolerant to grazing than prostrate species (white clover, perennial ryegrass). Italian ryegrass and red clover establish faster following seedling but are less persistent than fescues, perennial ryegrass, or white clover: they were still observed in 2015 but with low abundance, while fescues progressively overgrew Italian ryegrass or red clover after 3-4 years (Gastal *et al.*, 2012). No simple soil, climatic or management factor was strongly correlated with final botanical composition, but further investigation with multivariate analysis could identify combinations of factors explaining the current vegetation.

Conclusions

Initial results of the historical survey show that long-term grasslands can be found in a wide range of free-draining soil and oceanic climate conditions, with generally good persistence of sown species and the development of other common grazed grassland species, consistent with previous studies on ryegrass-white clover mixtures. Here, most old grasslands were sown with more than 2 species, which could be one important factor for persistence under a wider range of management practices. Combinations of species and management in the diverse soil and climate conditions should emerge; their contribution to ecosystem services will be assessed and compared to temporary grassland-arable rotations. Biodiversity already appears compatible with satisfactory production and forage quality (according to farmers), and longer grassland duration decreases the risks of carbon and N losses due to grassland destruction.

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Ploughing down and reseeding grassland on three dates in autumn: effect on crop yield and nitrate nitrogen concentration in the soil

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Abstract

Reseeding grassland in autumn is normally more productive in the first year than reseeding in spring, however it represents a higher risk for nitrate leaching. In an experiment in Merelbeke and Bocholt (Belgium), grassland was reseeded on 15 August, 15 September and 15 October in 2012. Evolution of the NO_3-N concentration in the soil and dry matter yields in 2012 and 2013 were measured to determine the best period of grassland reseeding for both the farmer (installation new sward, yield) and the environment (risk nitrate leaching). We found the best combination by reseeding in September. Reseeding in August resulted in high NO_3-N values in the soil in October-December and reseeding in October had a negative effect on sward development and yield in the year after reseeding.

Keywords: reseeding, yield, nitrate-N in soil

Introduction

Belgian dairy farmers prefer to reseed grassland in autumn rather than in spring for several agricultural reasons, but this timing has a negative impact on the environment because it represents a higher risk for nitrate leaching during autumn and winter. In an experiment in Merelbeke and Bocholt (Flanders, northern Belgium), grassland was reseeded in spring and autumn 2012. This paper describes the effects of timing of grassland reseeding in autumn on the NO_3-N concentration in the soil and the grass production capacity.

Materials and methods

Grassland was reseeded on 15 August (T1), 15 September (T2) and 15 October (T3) 2012 in Merelbeke (sandy loam, 50°59'NB 03°46'OL) and Bocholt (sandy soil, 51°10'NB 05°35'OL). Glyphosate (1,800 g ha^{-1}) was applied 2 weeks before and the 4-year original sward was ploughed down 1-2 days before sowing. Perennial ryegrass (*Lolium perenne* L.) was sown at a seed rate of 35 kg ha^{-1}. The trial design was a complete block design with 4 replicates. The field plots were 6×3 m; 6×1.5 m was harvested with a Haldrup forage harvester at a cutting height of 5 cm to calculate dry matter (DM) yield. In 2012 all autumn sown plots were harvested 5 times in the following year. DM yields of each were calculated after drying the subsample in a forced-draft oven at 80 °C for 24 h. Mineral N fertilization amounted to 300 kg N ha^{-1} for original sward and the perennial ryegrass sward in 2012 and 2013. The swards at T1, T2 and T3 before reseeding were cut 3, 4 and 4 times, respectively, in 2012. In 2013 all autumn sown plots were harvested 5 times. Soil samples were taken on all plots in 3 blocks and in 3 depth layers: 0-30 cm, 30-60 cm and 60-90 cm, with 6 drillings (drilling density on the mown strip: 33 drillings per 100 m^2). The soil samples were analysed for NO_3-N according to ISO 14256-2:2005.

Results and discussion

Effect on dry matter yield

Grassland is usually resown in September and so T2 is used as a reference to evaluate the effect of resowing time on the DM yield of the original sward in the year of reseeding and of the new sward the following year. Resowing in August (T1) had significant negative effects on DM yield of the original sward in 2012 at both locations. The difference was 1 cut of grass with a yield of about 2 ton DM ha^{-1} (Table 1). This difference in DM yield can vary greatly and depends largely on the quality of the original sward, the weather conditions and the N fertilizer use (Schils *et al.*, 2002). In this situation the original sward still had a good yield potential and the weather conditions were favourable for grass growth. Resowing in October (T3) had no significant effect on DM yield compared to T2 at either location. Difference in the time of sowing had a significant effect on the sward development, measured by the DM yield in the 1st cut, taken at the same time for T1, T2 and T3. The earlier the sowing in autumn, the higher was the DM yield in the 1st cut at both locations (Table 1). The low DM yield from T3 illustrated that the sward was not fully developed and tillering was not yet completed. T3 had a significantly lower annual DM yield in comparison with T2 and T1 at both Merelbeke and Bocholt, while the DM yield increase for T1 versus T2 was only significant at Merelbeke (Table 1).

Effect on the residual nitrate-N in the soil

The NO_3-N concentration increased significantly in comparison with the original sward, when the sward was ploughed down in every situation (time of resowing, location) and until the end of the year (Table 2). The measured NO_3-N concentrations after reseeding exceeded the threshold of 90 kg NO_3-N at 0-90 cm depth and there was a potential risk of leaching in the autumn and winter period (Flemish Legislation; https://www.vlm.be/nl/SiteCollectionDocuments/Mestbank/Algemeen/MAP5). The NO_3-N was significantly higher for T1 compared to T2 and T3 in 2012, but only remained far above the threshold of 90 kg until end of December in Merelbeke and until end of November in Bocholt. The NO_3-N for T2 and T3 was not significantly different in the period October-November. From December until April, T3 had a rather low but significantly higher nitrate-N concentration compared to T2. The low NO_3-N content in the soil in spring is not sensitive to leaching and is partly (upper layer) available for uptake by the young grass sward.

Table 1. Dry matter (DM) yield (t ha^{-1} and in %) of the original sward in 2012 and perennial ryegrass sown on 3 dates in autumn 2012 in 2013.

	DM yield, ton ha^{-1}			DM yield in %		
Glyphosate applied	1 Aug.	1 Sept.	1 Oct.	1 Aug.	1 Sept.	1 Oct.
Resowing after ploughing	15 Aug.	15 Sept.	15 Oct.	15 Aug.	15 Sept.	15 Oct.
	T1	T2	T3	T1	T2	T3
Merelbeke						
2012 sowing year[1]	11.4 a[2]	14.3 b	14.5 b	79.7 a[2]	100.0 b	101.5 b
2013 year after sowing						
1 cut	3.5 c	2.5 b	1.2 a	141.0 c	100.0 b	48.4 a
Total	13.1 c	11.9 b	10.8 a	109.7 c	100.0 b	90.9 a
Bocholt						
2012 sowing year	12.7 a	14.8 b	15.2 b	86.0 a	100.0 b	102.4 b
2013 year after sowing						
1 cut	3.9 c	2.0 b	0.7 a	191.6 c	100.0 b	32.8 a
Total	12.0 b	10.5 b	8.2 a	114.6 b	100.0 b	77.9 a

[1] DM-yield before resowing.

[2] Treatments with the same letter in the row are not significantly different.

Table 2. Nitrate-N in the soil profile 0-90 cm of the original sward and perennial ryegrass swards sown on 3 dates in autumn.

Location	Treatment		Nitrate-N in the soil profile 0-90 cm kg ha^{-1}						
			9/08/2012	**17/09/2012**	**16/10/2012**	**19/11/2012**	**17/12/2012**	**15/02/2013**	**29/03/2013**
Merelbeke	T0	Original sward	44 a[1]	41 a	28 a	19 a	12 a	10 a	11 a
sandy loam	T1	Sowing 15 August	57 a	49 b	255 c	190 c	138 d	25 b	12 a
	T2	Sowing 15 Sept.		53 c	105 b	114 b	89 b	31 b	17 a
	T3	Sowing October			40 a	107 b	109 c	49 c	49 b
			21/08/2012	**28/09/2012**	**30/10/2012**	**30/11/2012**	**28/12/2012**	**18/02/2013**	**2/04/2013**
Bocholt	T0	Original sward	121 a	170 a	120 a	71 a	19 b	13 ab	10 a
sandy	T1	Sowing 15 August	127 a	222 b	215 b	202 c	9 a	8 a	8 a
	T2	Sowing 15 Sept.		164 a	161 a	122 b	28 c	17 b	13 a
	T3	Sowing October			129 a	127 b	41 d	23 c	25 b

[1] Treatments within a location with the same letter in a column are not significantly different.

Conclusions

In this experiment reseeding grassland in August was the best for the establishment and the productivity of the new sward, but the level of NO_3-N in the soil was high from September until December because temperature and humidity were favourable for N-mineralisation, and there was thus a considerable risk of leaching. Reseeding grassland in October resulted in an incomplete establishment of the sward before the start of the new growing season and lower DM yields in the first year after sowing by comparison with reseeding in September. On the other hand, there were few significant differences between reseeding in October or September with respect to the NO_3-N content in the soil in 2012. Autumn reseeding of grassland in September was thus best in terms of benefits to both farmers and the environment.

Acknowledgements

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Ploughing down and reseeding grassland in April or at the end of May after one cut: effects on crop yield and nitrate nitrogen concentration in the soil

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Abstract

Reseeding grassland in spring is less productive in the first year than reseeding in autumn, but it is better for environmental reasons because it represents a lower risk for nitrate leaching. In experiments at Merelbeke and Bocholt (Belgium), grassland was reseeded either in April or at the end of May after one cut of grass. Evolution of NO_3-N in the soil and dry matter yield in the sowing year were measured to determine the best period of grassland reseeding both for the farmer and for the environment. In this experiment the dry matter yield of grassland reseeded on June 1st after one cut of the original sward was 0.9 t ha^{-1} higher in comparison with grassland reseeded on 15 April, but it was only significantly higher at one location and in one year. Moving the ploughing date from April to June 1st resulted in significantly higher NO_3-N concentrations in the soil during October – December 2012 and in October 2013, but the risk of nitrate leaching was still low.

Keywords: reseeding, spring, dry matter yield, soil NO_3-N

Introduction

Belgian dairy farmers prefer to reseed grassland in autumn rather than in spring for several agricultural reasons. If necessary, they also reseed grassland in spring: either in April or at the end of May after one cut of the original sward for silage. In experiments at Merelbeke and Bocholt (northern Belgium), grassland was reseeded in spring and autumn 2012 and in spring 2013. In this paper the effects of timing of reseeding grassland in spring on the NO_3-N concentration in the soil and on the grass production capacity are described.

Materials and methods

A 4-year old perennial ryegrass grass swards at two locations, Merelbeke (sandy loam, 50°59'NB 03°46'OL) and Bocholt (sandy soil, 51°10'NB 05°35'OL), were sprayed with glyphosate in both 2012 and 2013 (1,800 g ha^{-1} on 15 March (T1) or after 1 cut, i.e. 15 May (T3)). They were subsequently reseeded on 15 April (T2) or 1 June (T4), respectively (for further clarification, see Table 1). To prepare for sowing, the sward was ploughed down 2 days before sowing and the seedbed was prepared 1 day before sowing. Perennial ryegrass (*Lolium perenne* L.) was sown at 35 kg ha^{-1}. The trial design was a complete block design with 4 replicates. The field plots were 6×3 m, of which 6×1.5 m was harvested with a Haldrup forage harvester at a cutting height of 5 cm, to calculate dry matter (DM) yield. DM yields were calculated after drying the subsample in a forced draft oven at 80 °C for 24 hours. The mineral N fertilization was 300 kg N ha^{-1} $year^{-1}$ in 2012 and 2013, except in the year of reseeding when it was 220 kg N ha^{-1} for reseeding at T1 and T3 and 200 kg N ha^{-1} for reseeding at T2 and T4. The reseeded swards were cut 4 times in 2012 and 3 times in 2013 on both locations. Soil samples were taken on all plots in 3 blocks at 3 depths: 0-30 cm, 30-60 cm and 60-90 cm, with 6 drillings (drilling density on the mown strip: 33 drillings per 100 m^2). Soil samples were taken before or at least 2 weeks after the application of N-fertilizer. The sampling methodology was the same for each plot and each time of sampling. The soil samples were analysed for NO_3-N according to ISO 14256-2:2005.

Results and discussion

Effect on dry matter yield

Grassland is usually reseeded in April but sometimes farmers prefer to harvest one fertilized cut from the old sward in the beginning of May, sometimes with an application of glyphosate in mid-May and subsequent reseeding at the end of the month. Thereby they harvest a high quality first cut in spring, and obtain a good glyphosate uptake and effect and a good soil temperature for germination and emergence of the new seedlings. The average DM yield for reseeding at T1, T3 (April 15th) and T2, T4 (June 1st) in 2012 and 2013 for the 2 locations, was 9.7 t ha^{-1} and 10.8 t ha^{-1}, respectively, but the yield increase was only significant at Merelbeke in 2012 (Table 1). The cut before reseeding on 1 June was fertilized with 100 kg $N_{available}$ ha^{-1} and the average DM yield was 3.6 t ha^{-1} (34% of the total annual DM yield). This yield depended on the botanical quality of the sward, N fertilization, the weather conditions and the cutting date. The DM yield of the reseeded grassland in the year of reseeding was significantly lower in comparison with that of the original sward, which was still of good quality after 4 years of cutting management, after receiving an extra N fertilization of 80-100 kg N ha^{-1} and producing an extra cut.

Effect on the residual NO3-N in the soil

When the time of ploughing down the sward was shifted from 15 April to 1 June, the N mineralization started later and this resulted in significantly higher NO_3-N concentrations in the soil in October – December 2012 and in October 2013 at Merelbeke (Figure 1). The results of Bocholt (not shown here)

Table 1. Dry matter (DM) yield (t ha^{-1}) of the swards reseeded in April or June in 2012 and 2013.[1]

		DM, t ha^{-1}			
Glyphosate appl.	no	15 March 2012	15 May 2012	15 March 2013	15 May 2013
Date resowing	no	15 April 2012	1 June 2012	15 April 2013	1 June 2013
Merelbeke					
2012	15.2 (5)[2] b	9.6 (4) a	12.0 (4) b		
2013	12.0 (4) b			8.6 (3) a	8.8 (3) a
Bocholt					
2012	15.8 (5) b	13.0 (4) a	14.4 (4) a		
2013	8.4 (4) a			7.5 (3) a	8. 0 (3) a

[1] Treatments with the same letter in a row are not significantly different *P*<0.05.

[2] (x), x is number of cuts.

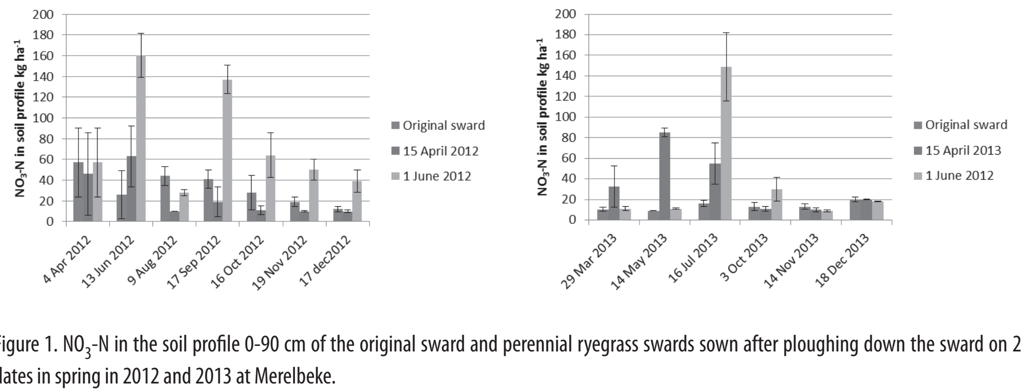

Figure 1. NO_3-N in the soil profile 0-90 cm of the original sward and perennial ryegrass swards sown after ploughing down the sward on 2 dates in spring in 2012 and 2013 at Merelbeke.

confirmed these measurements. The measured NO_3-N after reseeding in April and at the end of May was nevertheless considerably below the threshold of 90 kg NO_3-N at 0-90 cm soil depth (https://www.vlm.be/nl/SiteCollectionDocuments/Mestbank/Algemeen/MAP5), and as a result the risk of NO_3-N leaching in the autumn and winter period was low. These results are confirmed by the study of Velthof and Hummelink (2012) on sandy soils in 2010 and 2011, where they reported that mineral N content of the soil in November showed only small differences between reseeding in April, end of May and end of June and the risk of N leaching did not increase when ploughing down and reseeding was delayed from April until July 1st.

Conclusions

In this experiment the DM yield of grassland reseeded on June 1st after one cut of the original sward was 0.9 t ha^{-1} higher in comparison with grassland reseeded on 15 April, but it was only significantly higher at one location and in one year. The cut before reseeding on 1 June was fertilized with 100 kg $N_{available}$ ha^{-1} and the average DM yield was 34% of the total annual DM yield. Moving the date of ploughing down the sward from 15 April to 1 June resulted in significantly higher NO_3-N concentrations in the soil during October – December 2012 and in October 2013, but the risk of nitrate leaching was low.

Acknowledgements

We wish to thank PVL Bocholt for their cooperation and the Agricultural Centre for Forage Crops and the Advisory Service of the Flemish Government for funding.

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Measuring root systems in forage legumes: a comparison of two systems

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Abstract

This study compares two systems of measuring root amount and distribution in 11 forage legume populations: machine scanning and manual measurement. The methods produced similar results, and advantages and disadvantages of both are discussed.

Keywords: forage legumes, phenomics, roots

Introduction

The current emphasis in forage breeding on delivery of environmental benefit as well as high-quality biomass requires plants to capture water and nutrients efficiently, and to enhance soil quality. Greater rooting depth, together with an increased proportion of root biomass lower down the root profile, have been proposed as useful traits in this respect due to the positive effects of plant roots on soil structure (Macleod *et al.*, 2007). Gains in traits of agronomic value in forages, such as shoot biomass production, have not always resulted in concomitant improvements in root mass (Crush *et al.*, 2010), so there is a need for breeding programmes to address this. Technical difficulties associated with assessing plant root systems have impeded investigation of their potential to deliver environmental benefits. A five-year UK project, 'Sureroot', focuses on this deficiency in knowledge, and measures the impacts of root system architecture on soil quality and hydrology. Work on forage legumes in Sureroot focuses on three crops: red clover (*Trifolium pratense*), white clover (*Trifolium repens*) and a hybrid between white clover and the rhizomatous species Caucasian clover (*Trifolium ambiguum*). This study compares results from analyses of root distribution carried out on 11 forage legume populations grown in either perspex containers ('root columns') and analysed by machine scanning, or in plastic cylinders ('root pipes') and measured manually.

Materials and methods

Root growth was measured in the following populations: red clover commercial varieties AberChianti, AberClaret, Britta, Milvus and advanced breeding line Aa 4559; white clover stolon mapping family populations 'Thick Profuse' and 'Thin Sparse' (Collins *et al.*, 1997), commercial varieties AberAce, AberDai, Aran, and hybrid variety AberLasting, which exhibits the rhizomatous trait. An experiment was set up in autumn 2014 in which plants were grown under glasshouse conditions (minimum temp 20 °C/10 °C day/night; minimum day length 10 hours) in a replicated, randomised design for measurement of root traits using two techniques: (1) 10 plants of each population were individually established in transparent perspex containers ('root columns': 11×11×50 cm depth) which were scanned by machine in the National Plant Phenomics Centre (NPPC) in IBERS, during which images of root columns were processed automatically to deliver proxy measurements of root mass; (2) 10 plants of each population were individually established in plastic cylinders ('root pipes': 17.8 cm diam. × 100 cm depth). Within each cylinder the plant was encased in a clear polythene sleeve which was lifted out for manual measurement of root growth made by counting the number of roots intersecting horizontal lines drawn at a series of soil depths (5, 10, 50 and 80 cm). In both systems plants were grown in Levington's F2 compost – a fine structured, free flowing growth medium. A layer of gravel 5 cm deep was added to the base of each container to allow water drainage via small drilled holes. All plants were inoculated with an

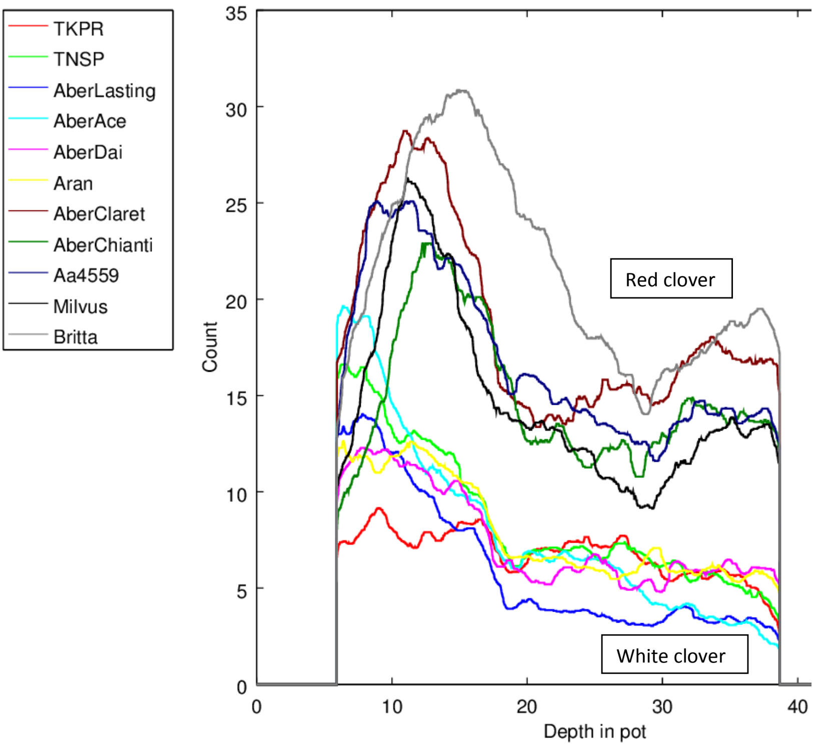

Figure 1. Root count (a proxy for root mass) scanned at depths in the root profile in plants of red and white clover populations grown in perspex columns.

appropriate *Rhizobium* culture and a high level of nodulation was observed. Measurements were made at monthly intervals during spring-early summer 2015, and stopped when roots had grown to the bottom of the containers. A total of five 'root column' and three 'root pipe' root measurements were made.

Results and discussion

The two measurement systems produced broadly similar results for distribution of biomass down the root profile. Differences between the legume species became more apparent over time, so here we present results from later in the experiment. Figure 1 shows results from the NPPC scan of early June 2015. The response for each population was derived from the four faces of the perspex column containing each genotype, and aggregated over10 genotypes. Thus, imprecisions in the method were compensated for by averaging over multiple views.

The two species can be clearly distinguished, with red clover producing greater root mass than white clover at most depths and showing a more even pattern of distribution down the profile. In contrast, root mass in white clover tended to be located higher in the profile, at depths <20 cm. Within white clover, the *T. ambiguum* hybrid, AberLasting, behaved in a way that was typical of other white clover populations in terms of root distribution but had somewhat less root mass at depths <20 cm. The advantage of using machine scanning to measure root distribution is that the very large amount of data generated is ideal for implementing mathematical curve fitting procedures – these could be used to compare the pattern and dynamics of root distribution within and between populations. On the other hand, the dimensions of the perspex columns were a constraint to root growth and no measurements could be made at depths >40 cm. In addition, the NPPC facility is expensive to operate. In contrast, 'root pipe' measurements were inexpensive to set up but laborious to carry out. Figure 2 shows results from the 'root pipe' measurement in late May 2015. The species differed in terms of the overall size of the root system, and in the root mass present at each depth in the profile (i.e. the pattern of root distribution).

There was a significant difference between populations (P<0.001) in total root mass (the sum of root mass at each depth), and red clover populations produced much bigger root systems. The population with the largest root system was red clover Britta (345 intersections), compared with 96 intersections in the population with the smallest, white clover AberAce (standard error of the difference (sed) = 33). There were significant differences between populations at all depths down the root profile. The white clover AberDai had the highest root mass in the top 5cm of the profile (79 intersections) and the red clover AberClaret the lowest (40 intersections) (sed = 10). Differences between the species were less evident at 10 cm depth, but at depths >50 cm red clover produced significantly more root mass. At this depth Britta produced by far the most root mass amongst the red clover group. In conclusion, this ecologically important trait requires measurement in suitably deep containers, so the 'root pipe' system appears superior in this respect, as roots could be measured to a depth of about 80 cm. However, root distribution measurements <50 cm were similar in the two systems.

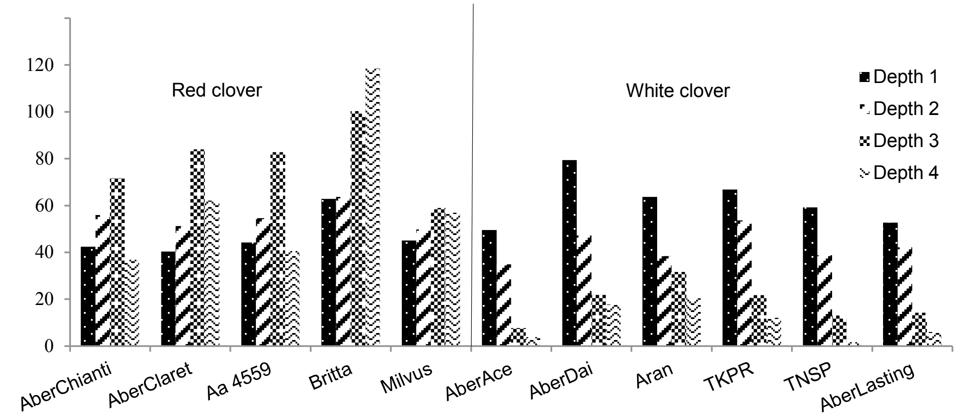

Figure 2. Number of root intersections (a proxy for root mass) with horizontal lines drawn at four depths in the root profile: 1 = 5 cm; 2 = 10 cm; 3= 50 cm; 4 = 80 cm. Intersections were counted by eye. Plants were grown in root pipes.

Acknowledgements

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Impact of ecological factors on the flooded meadow phytocoenosis structure and productivity

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Abstract

A field trial was conducted in flooded meadows on different parts of the Nemunas (the biggest river of Lithuania) delta in 2009-2012. The mixture of the grass was sown in 1985 in this experimental territory. After 1992, the meadows were not fertilized and hay harvesting was done only once. The study includes data of the changes of botanical composition, stability of phytocoenosis, dry matter yield, economic value with the aim of finding relation between these parameters and different soil agrochemical characteristics, groundwater level, flood period duration. The greatest plant species diversity, with the greatest economic value (56.9±1.54) was established in flooded meadow on the riverside part of the Nemunas delta behind a levee (flood period is short: 10-20 days) in Areni Calcicaric Fluvisol (Flc-ar) soil, with low acidity, medium nitrogen content, rich in phosphorus and potassium. The biggest dry matter yield (8.64±0.77 t ha^{-1}), but the least economic value of meadow (41.1±3.41) and the least diversity of plant species was found in the central part of the Nemunas delta (flood period long – up to 2 months) in Terric Histosol (HSs) soil, with moderate acidity, rich in nitrogen, but low in potassium and phosphorus.

Keywords: flooded meadow, botanical composition, yield

Introduction

Nemunas delta region (Western Lithuania) is characterized by flooded meadows with a great variety of flora and high dry matter yield. Most of these meadows are currently under extensive management. Floristic changes, as an effect of semi-natural meadow gradual extensification, has been frequently reported (Katutis, 2008). Previous results indicated a relationship between plant species composition diversity and ecological factors and management character (Gusmeroli *et al.*, 2013). According to Huyghe *et al.* (2008), dry matter yield is related to the phytocoenosis structure and the physiology and morphology of the plants. This in turn affects the feeding value. However, there is still a lack of information on the direction of vegetation changes, dry matter yield, depending on soil agrochemical parameters and humidity regimes. The aim of this study was to estimate the influence of humidity regime and soil agrochemical parameters on the flooded meadow phytocoenosis structure and productivity.

Materials and methods

The research was carried out during the period 2009-2012 in three flooded meadows, in different parts of the Nemunas delta, Western Lithuania: in the riverside (soil = Areni calcicaric Fluvisols (Flc-ar); 170 ha), central (soil = Terric Histosols (PDp 1); 380 ha) and pre-land (soil = Endohipogleyi Fluvisols Flc-gln-w; 100 ha). The mixture of the grass consisted of *Phleum pratense* L. 15%, *Poa pratensis* L. 10%, *Poa palustris* L. or *Agrostis stolonifera* L. 15%, *Alopecurus pratensis* L. 30%, *Typhoides arundinacea* (L.) Moench 30% was sown in 1985. Until 1992, the meadows were fertilized with $N_{70\text{-}90}P_{30\text{-}40}K_{70\text{-}90}$ and cut 4-5 times. After 1992, the meadows were not fertilized and hay harvesting was done once during the vegetation period. There were different agrochemical characteristics and groundwater levels in the three experimental sites (Table 1).

Table 1. Agrochemical characteristics of the soil of flooded meadows on different parts of the Nemunas delta at the beginning of the experiment (2009) and fluctuation in the groundwater level (GWL) during the plant vegetation (cm).

Part of the Nemunas delta	Agrochemical characteristics					GWL fluctuation in 2009-2012, cm. (min/max)			
	pH_{KCl}	N_{total} (%)	Mobile		$C_{organic}$ (%)	2009	2010	2011	2012
			P_2O_5 (mg kg^{-1})	K_2O (mg kg^{-1})					
Riverside	6.9	0.261	154	163	2.530	-150 and over			
Central	5.9	0.388	59	96	3.687	-42/+2	-31/+12	-75 /0	-60/+37
Pre-land	5.5	0.221	111	83	2.340	-143/-56	-118/-5	-145/-58	-135/+27

Plant composition assessment was performed according to De Vries method (Peeters, 1989). Depending on the meadow area, research was carried out at 3 or 5 experimental sites, each with an area of 100 m^2. Dry matter yield was determined by cutting 1 m^2 plots once per year (six replications at each site). Calculation of Shannon-Wiener Index H' was made to estimate the changes in the structure of meadow phytocoenosis (Villanueva-Rivera *et al.*, 2011):

$$H' = -\sum (Pi \times \ln Pi)$$

Where Pi = proportional abundance of the ith taxa.

The meadow's economic value was calculated by multiplying the relative abundance of each grass species by its respective economic value: on a points scale from 0 to100 (Peeters, 1989).

Results and discussion

The greatest diversity of plant species (35-53 species in different year) was identified in the riverside part of the Nemunas delta meadow. The grass sown 27 years ago almost perished, especially *T. arundinacea* (L.) Moench and *A. pratensis* L. The greatest part of *Fabaceae* plant family was determined compared to the plant variety that is typical to this family in other meadows. *A. stolonifera* L., *Dactylis glomerata* L., *Festuca rubra* L., *Lathyrus pratensis* L. can be partly treated as species that form phytocoenosis. Shannon-Wiener Index (H') indicated quite great stability of this phytocoenosis (H'=3.050-3.469), but dry matter (DM) yield was relatively low (from 2.87 to 7.20 t ha^{-1}) (Figure 1). However, the economic value of this phytocoenosis was the biggest: 56.9±1.54. In the central part of the Nemunas delta, the least variety of plant species was established (12-18 species in different year). In this location plants of hydrofits ecological group dominated: *Carex* spp., *Iris pseudocorus* L., *Galium palustre* L., *Rorippa palustris* L. Besser, *Sium latifolium* L. Grasses composed approx. 60% of all biodiversity. Grass species with higher economic value were not found, so this value was the lowest (41.1±3.41), although the DM yield was the highest (from 7.09 to 10.55 t ha^{-1}) in comparison with other meadows (Fig. 1). Large fertility potential of the central part of the Nemunas delta was influenced by a favourable humidity regime and soil rich in nutrition. DM yield depended on the common number of species and abundance of *Poaceae* family plants. Significant differences in DM yield were observed between all meadows. In the pre-land part of the Nemunas delta meadow, affected by short-term floods, a phytocoenosis was formed with a moderately rich number of species (26-37 species). According to its structure and DM yield, this phytocoenosis is quite similar to those formed in the riverside part of the Nemunas delta (H'=2.816-3.256). During the investigative period there was no dominant establishment common for the whole association. *A. pratensis* L., *P. pratensis* L., *Stellaria palustris* Retz., *Veronica longifolia* L. can be treated as species that form phytocoenosis.

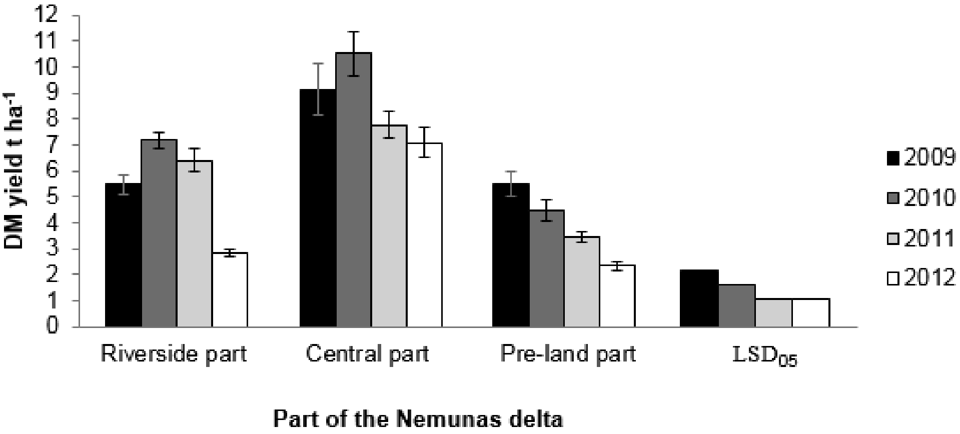

Figure 1. Dry matter (DM) yield of meadow phytocoenosis (t ha^{-1} ± standard error) on different parts of the Nemunas delta, Western Lithuania, 2009-2012.

Conclusions

Botanical composition of meadows depended on agrochemical characteristics of the soil: the greatest plant species diversity, and the best phytocoenosis stability was established in Areni Calcicaric Fluvisol (Flc-ar) soil, with low acidity, and rich in phosphorus and potassium. Humidity regimes, GWL ranges during the plant vegetation period, and nitrogen amount in the soil all influenced the DM yield of meadow phytocoesosis. There were no relations between DM yield and plant species number, but DM yield depended on the abundance of *Poaceae* family plants. The economic value depended on prevailing plant species.

Acknowledgments

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Does the variety of *Lolium perenne* affect the performance of binary and multi-species mixtures?

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Abstract

The benefits of growing *Lolium perenne* and *Trifolium repens* in mixtures have long been recognized. Increasing interest has recently been given to the productivity of multi-species mixtures including forbs. Little is known of whether genetic variation within forage species affects competition and complementarity among species in mixed stands and therewith overall sward performance. An experiment was performed with four varieties of *L. perenne* grown either in monoculture with nitrogen fertilization (200 kg ha^{-1} a^{-1}) or in unfertilized binary (with *T. repens*) and multi-species mixtures (including *Plantago lanceolata* L. and *Taraxacum officinale* Web.). The *L. perenne* varieties differed in time of heading (early, late) and growth habit (prostrate, erect). Aboveground herbage was cut four times a year over four years. The results showed that the unfertilized multi-species mixture had the same total yield as the fertilized *L. perenne* monoculture. The erect form of *L. perenne* showed a significantly higher yield than the prostrate one, both in *L. perenne* monoculture and in mixture with *T. repens*. In the multi-species mixture the *L. perenne* variety had no effect on the total dry matter yield.

Keywords: multi-species swards, perennial ryegrass, forbs, white clover, species compatibility

Introduction

In recent years, increasing attention has been paid to binary mixtures of *Lolium perenne* and *Trifolium repens* and also to multi-species mixtures. Little evidence exists that the variety within a forage species would affect the mixture yield. The inclusion of a forb as a third component in a grass-clover mixture may provide production advantages (Assaf and Isselstein, 2009). So far, no information is available whether the choice of the grass variety might affect the performance of a multi-species mixture. An experiment was therefore established using contrasting grass varieties of *L. perenne* grown in binary mixtures with *T. repens* and multi-species mixtures with the forbs *Plantago lanceolata* and *Taraxacum officinale*, both of which are widespread in grasslands all around the world (Labreveux *et al.*, 2006).

Materials and methods

The field experiment was implemented on a well-drained, fertile luvisol at the Experimental Station Reinshof of the University of Goettingen, Germany. Four *L. perenne* varieties were each sown in different stands: monocultures, binary mixtures with *T. repens* (0.75 : 0.25) and in multi-species mixtures with *T. repens*, *P. lanceolata* and *T. officinale* (0.4 : 0.2 : 0.2 : 0.2, proportions based on number of germinable seeds m^{-2}). Sowing density in all stands was 2,000 germinable seeds m^{-2}. The varieties of *L. perenne* were chosen to represent factorial combinations of traits, namely phenology (early and late heading) and growth habit (prostrate, erect). Treatments were set up in a randomized block design with four replications. Herbage was harvested 4 times per year over four years. Subsamples of fresh herbage were hand-separated into *L. perenne*, *T. repens*, *P. lanceolata* and *T. officinale*, and non-sown species, and dried to determine the component yields. The *L. perenne* monoculture was fertilized with 200 kg N ha^{-1} per year. No phosphorus or potassium fertilizer was applied. Extractable (calcium acetate lactate) soil nutrient concentrations at the end of the experiment were 90 mg kg^{-1} phosphorus and 130 mg kg^{-1} potassium, with a pH of 5.9 (CaCl). The total dry matter yield and each component yield were analysed by Linear Mixed-Effects

Models (Lme). Model reduction was performed and multiple comparison test (LSD) using the software R.3.2.3 (R core team, 2015) and the packages 'nlme' (Pinheiro *et al.*, 2016) and 'lsmeans' (Russell, 2016).

Results

There was a significant interaction growth form × stand in the cumulative total dry matter yield (Table 1). Highest yields were obtained by the fertilized pure *L. perenne* sward of the erect growth form. The yield of the erect growth form was higher than the prostrate form. However, this effect was diminished in the binary mixture and even more so in the multi-species mixture. There was no significant difference between the grass monoculture and the multi-species mixture when the erect growth form of *L. perenne* was used. Referring to prostrate growth form, the total dry matter yield of the multi-species sward mixture was higher than of the monoculture.

The *L. perenne* dry matter yield was significantly affected by stands and growth form (Table 1). The highest *L. perenne* dry matter yield was found in pure stand and the erect form was higher yielding than the prostrate form. The yield of the other components (*T. repens*, *P. lanceolata, T. officinale* and the weeds) were neither affected by the factor stand nor by the grass variety. In addition, the interaction of cultivation × growth form was not significant.

Discussion and conclusions

This study confirms that the variety of *L. perenne* may strongly affect the herbage yield of monocultures. However, in binary mixtures with *T. repens* and multi-species mixtures no significant difference among the different varieties was found. It seems that the more species that are present in a forage mixture, the lower is the importance of varietal variation within the grass partner. This might be explained by the broad range of plant traits in multi-species mixtures, such as growth habit, growth rate or rooting system, which allow for a complementary resource use (Assaf and Isselstein, 2009), and which then leave little room for further complementarity effects by grass varieties.

Table 1. Cumulative dry matter (DM) yields over four years. *Lolium perenne* varieties differing in growth form (erect = E, prostrate = P) cultivated as monocultures (G), binary mixtures (G/C) and multi-species mixtures (G/C/F).[1,2]

		Cumulative DM yield (kg ha^{-1})			Growth form	Phenology	Stand	Growth form × Stand
Yield	**Growth form**	**G**	**G/C**	**G/C/F**	***P*-values**			
L. perenne	E	27,193aA	13,209aB	7100aC	<0.0001**	–	<0.0001**	–
	P	22,207bA	10,877bB	4469bC				
T. repens	E	0	10,860	11509	0.273^{NS}	0.749^{NS}	0.780^{NS}	0.774^{NS}
	P	0	11,887	12293				
P. lanceolata and	E	0	0	8547	0.1^{NS}	–	–	–
T. officinale	P	0	0	9664				
Weed	E	603	649	486	0.692^{NS}	–	0.745^{NS}	0.774^{NS}
	P	661	679	695				
Total	E	28,451a	25,039bc	27862ab	0.003**	–	0.007**	0.040*
	P	23,232c	23,748c	27337ab				

[1] *T. repens* was not present in the G sward, *P. lanceolata* and *T. officinale* not in the G/C mixture.

[2] Total dry matter yield data indicated by the same character are not significantly different. For *L. perenne* small letters indicate differences among growth form and capital letters among stands. NS = not significant.

As has been found earlier, our results showed the potential benefits of multi-species mixture for herbage production compared to binary grass-clover mixtures. More research is needed to establish to what extent this is due to the particular traits of the sown grassland forbs and whether this information could be used to design highly efficient grass swards.

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Aboveground-belowground biodiversity linkages in dairy sheep systems with different grazing regimes

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Abstract

Understanding linkages between plant diversity aboveground and that of microorganisms belowground constitutes an important challenge for the sustainability of permanent pastures and the knowledge of the ecosystem services that they provide. This study was carried out to test the effect of two different grazing regimes (called regenerative grazing and free grazing) and ungrazed treatment at pastures' biodiversity level. Data collected after a one year grazing regime were used to search for the linkages between plant and microbial communities. According to the results, plant richness and belowground biodiversity showed a tendency towards lower values under ungrazed conditions, and both indexes showed a tendency to higher diversity in a regenerative grazing regime. Relating to the interactions between aboveground plant structural and belowground soil functional composition, bare ground had a weaker correlation with the catabolic potential of soil microorganisms.

Keywords: agroecosystem health cards, plant diversity, rotational grazing, soil microbial diversity

Introduction

The main objective of Life Regen Farming (www.regenfarming.eu) project is to demonstrate the viability of regenerative agriculture practices and their technic-economical and environmental effectiveness. One of the tested practices is the grazing regime and its effect on soil and vegetation parameters. There is an extended literature concerning the influence of sheep grazing on plant species composition and productivity (Jing *et al.*, 2014; Ren *et al.*, 2012). Besides, it is known that soil microbial communities influence plant community composition (Barrios, 2007). However, the influence of grazing in the interaction between plant and soil microbial communities remains unexplored. The objective of the study was to determine the effect of different grazing regimes (regenerative, free or ungrazing) on aboveground-belowground biodiversity relationships: plant diversity, soil diversity, and correlations between above- and belowground.

Materials and methods

An experiment was conducted with the experimental flock of NEIKER-Tecnalia in spring 2015. Sheep were blocked into two homogeneous groups of 60 head each and randomly assigned to regenerative (RG) or free (FG) grazing regimes under the same stocking rate. Grazing and resting days of each gazing regime are detailed in Mandaluniz *et al.* (2015). As a third treatment (ungrazed), exclusion fences (3×3 m^2) were built at the 3 paddocks, in order to monitor the effect of grazing abandonment on aboveground and belowground biodiversity.

Soil samples and plant data were collected after one year of managing the sward under the different grazing regimes. At the aboveground level, plant richness (S) and Shannon's (H') diversity index were calculated by Estimates software (Colwell, 2006). This sampling was carried out at fixed transects, measuring 10 squares per paddocck (0.5×0.5 m^2). At the belowground level, soil samples were collected near the fixed transects. Samples were processed and community level physiological profiles of bacteria were studied using Biolog EcoPlates^{TM} (Epelde *et al.*, 2008). The plates corresponding to an incubation time of 48 h were chosen for diversity indexes calculations. The number of utilized substrates (i.e. the number of substrates with an absorbance value >0.25; this value marked the beginning of the exponential phase in the Biolog EcoPlatesTM), equivalent to S index was calculated at this 48 h incubation. Similarly, H' index was calculated considering absorbance values at each well as equivalent to species abundance.

All data were analysed by R software considering as fixed effect the grazing management (RG, FG and exclusions). Pearson correlation values for S and H´ between plant and soil microbial communities were also calculated and a heat-plot of the correlation matrix between plant structural composition and microbial functional composition was elaborated. The reordering of plant species and carbon substrates was performed according to the similarity of the correlation values.

Results and discussion

At the aboveground level, the overall mean plat richness (S) was 13±2 and Shannon's diversity index (H´) was 1.5±0.3, considering the paddock as a random variable. Plant richness was slightly lower under exclusion treatments (Table 1). This result shows the effectiveness of grazing on the maintenance of plant biodiversity in these pastures (Hill *et al.*, 1992). On the other hand, no differences were observed among grazing treatments (FG vs RG). At the belowground level, mean soil microbial community's richness (S) was 23±2 and Shannon's diversity index (H´) was 4.3±0.2. According to an ANOVA statistical analysis, no differences were observed among treatments regarding functional richness or diversity index, but both indexes showed a tendency of lower diversity at the exclusion areas (Table 1).

Relating to the correlation between the aboveground and belowground biodiversity indexes, non-conclusive results were observed (Table 1), but a negative tendency was observed at exclusion (P=0.078). In this sense, there is a wide literature regarding the positive interaction between aboveground plant diversity and belowground microbial communities, which significantly contributes to ecosystem functioning (Bardgett and Van der Putten, 2014).

Regarding the interactions between aboveground plant structural and belowground soil functional composition, it was remarkable that bare soil had a weaker correlation with the substrate utilization potential of soil microorganisms. Finally, plant species with higher cover of the studied paddocks (e.g. *Lolium multiflorum, Festuca arundinacea*) were the ones that showed a stronger correlation with the potential substrate utilization of soil microorganisms. The study of these interactions would be an important step towards the sustainability of these pasture-based systems and to the knowledge of the ecosystem services that they provide. This monitoring will go on under the project Life Regen Farming, to confirm these tendencies at a midterm period.

Table 1. Plant and microbial functional richness (S) and Shannon's (H´) diversity index (average ± standard deviation, n=3 replicates). Pearson correlation values for S and H´.

		Plant communities	Soil microbial communities	Pearson Correlation	Significance
Regenerative grazing	S	14±2	23±2	-0.126	0.920
	H´	1.6±0.1	4.3±0.2	0.760	0.450
Free grazing	S	14±0	23±2	–	–
	H´	1.5±0.2	4.3±0.1	0.062	0.960
Ungrazed	S	12±4	22±1	-0.993	0.078
	H´	1.5±0.5	4.2±0.1	-0.972	0.150

Conclusions

According to the results, plant richness and belowground biodiversity are affected by the grazing activity. In general these values are lower under ungrazed conditions. Moreover, grazing regime influenced the correlations between richness and diversity indexes calculated for plant and microbial communities.

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Litter decomposition on a heterogeneous cattle pasture is influenced by sward structure

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Abstract

Pastures with low grazing pressure are often characterized by a sward mosaic of short, frequently grazed and tall, rarely grazed patches. These two patch types differ in the quantity of litter returned to the system, but may also produce different litter quality and provide different conditions for decomposition. We collected litter from short and tall patches of a cattle pasture in both May and August. A litter bag method was used to assess *in situ* decomposition of these litter samples in either short or tall patches over a period of two to six weeks after sampling. We expected faster decomposition of litter from short patches and of litter incubated in tall patches. Contrary to our first hypothesis, litter from short patches only showed faster decomposition than litter from tall patches during the second incubation period. During the first incubation period, the opposite was the case. Compared to litter incubated in short patches, litter incubated in tall patches tended to decompose faster, confirming our second hypothesis.

Keywords: litter decomposition, patch grazing, heterogeneity, nutrient cycling

Introduction

In grazing systems with low grazing pressure, selective grazing of animals often results in a heterogeneous sward structure consisting of short and tall patches through a process called 'patch grazing' (Dumont *et al.*, 2012). Short patches are defoliated frequently so that a large proportion of the plant mineral nutrients is returned to the soil via animal urine and dung. In the rarely defoliated tall patches, nutrients largely enter the soil through the litter cycle. These differences should affect the overall speed of nutrient cycling in each patch type, since the excrement cycle is generally assumed to be faster than the litter cycle (Ritchie *et al.,* 1998). Additionally, the speed of the litter cycle itself is affected by grazing intensity: In productive systems, grazing generally increases the competitiveness of productive species with high nitrogen concentration, that produce easily degradable litter (Güsewell *et al.*, 2005). This would speed up litter decomposition in short patches, accelerating the speed of nutrient cycling and potentially the productivity of these patches even further.

We studied sward-structure effects on litter decomposition in a low-intensity cattle pasture by incubating litter from two contrasting patch types either in the same or in the opposite patch type. We assumed that litter from short patches would decompose faster, as we expected these patches to contain plants with higher nitrogen concentration. We also expected litter decomposition to be faster when both types of litter were placed in tall patches, due to a more favourable microclimate close to the soil surface, but that this effect would be subordinate to that of litter origin.

Materials and methods

The study was conducted on a long-term cattle grazing experiment established in 2002 in Relliehausen, near Goettingen (51.78 °N, 9.70 °E), on a moderately species-rich *Lolio-Cynosuretum*. Three 1 ha-paddocks had been grazed to a target compressed sward height of 12 cm by Simmental suckler cows in a continuous stocking system. The pasture was neither mown nor fertilized during the experiment. In each paddock, aboveground biomass was collected from short and tall patches two times (May 6 and August

4-5 2015, respectively) by stratified random sampling. Collected biomass was separated into living and dead biomass ('litter'). Litter samples were analysed for their carbon (C) and nitrogen (N) concentrations using an elemental analyser. A mixed litter sample across paddocks was produced for both short and tall patches at each date to fill litter bags. Litter bags had a mesh width of 1 mm and were filled with 3 g each of air dried litter. They were incubated on the experimental site under temporal exclusion cages, starting either June 2 (summer incubation) or August 25-26 (autumn incubation). Per paddock, two short and two tall patches were selected as 'incubation patches'. Per incubation patch, three litter bags of each 'litter patch type' (short/tall) were placed directly on the soil surface after removing aboveground vegetation and directly under the litter bag, but leaving surrounding vegetation intact. Two, four and six weeks after the start of the incubation one litter bag per litter patch type and incubation patch was collected. The biomass contained inside was dried and weighed. Its organic matter (OM) concentration and that of the original mixed litter samples were determined by loss on ignition at 550 °C. Relative OM loss during incubation was calculated as the difference between initial OM mass and OM mass collected after incubation, divided by initial OM mass. Mean temperature generally increased during the summer incubation period with values of 15.9, 13.8 and 18.1 °C during the first, second and third fortnight of the summer period, while it decreased during the autumn incubation period (15.7, 13.2, 10.7 °C). Total precipitation during each fortnight followed the same pattern, with 2, 29, 73 mm in summer and 49, 30, 5 mm in autumn.

Linear mixed effects models were fitted to predict relative OM loss from the fixed effects litter patch type, incubation patch type, incubation duration, season, and their interactions. C:N ratio of litter was modelled likewise, with litter patch type, season and their interaction as fixed effects. Minimum adequate models were determined based on Akaike's Information Criterion. Tukey tests were performed to compare means in case of significant effects. The software R (R Core Team, 2015) and the packages 'nlme' (Pinheiro *et al.*, 2015) and 'lsmeans' (Lenth and Hervé, 2015) were used for the analysis.

Results and discussion

With increasing incubation time, there was a general increase of litter OM loss, with only minor modifications through interactions with litter patch type, incubation patch type, or season (Figure 1). Differences in average decomposition rates between summer and autumn and between the three incubation durations reflected the differences in temperature and precipitation. As expected, litter decomposition tended to be faster under incubation in tall patches, although this effect was only significant for the four-week incubation. Litter patch type, however, had a more complex effect on litter decomposition: In autumn, litter from short patches decomposed faster than litter from tall patches, as hypothesized. In summer, the opposite was true: Litter from tall patches showed higher OM loss than litter from short patches (Figure 1).

This interaction could only partly be explained by litter C:N ratio. In spite of a trend towards larger values in tall patches, neither patch type (P=0.166) nor its interaction with sampling date (P=0.182) had a significant effect on the C:N ratio. Irrespective of patch type, litter C:N ratio was smaller (P=0.020) in May (28.4±1.4 and 29.9±3.9) than in August (43.6±11.2 and 61.5±18.3; mean ± standard deviation of short and tall patches, respectively). The considerable variation of litter C:N ratios between blocks in autumn was, however, levelled in the incubated samples which constituted a mixture of all three blocks. Consequently, decomposition of litter from tall patches may have been limited by N concentration in autumn. The effects of other litter quality parameters, such as lignin concentration, or seasonal changes in the activity of different decomposers are further possible explanations for the observed result.

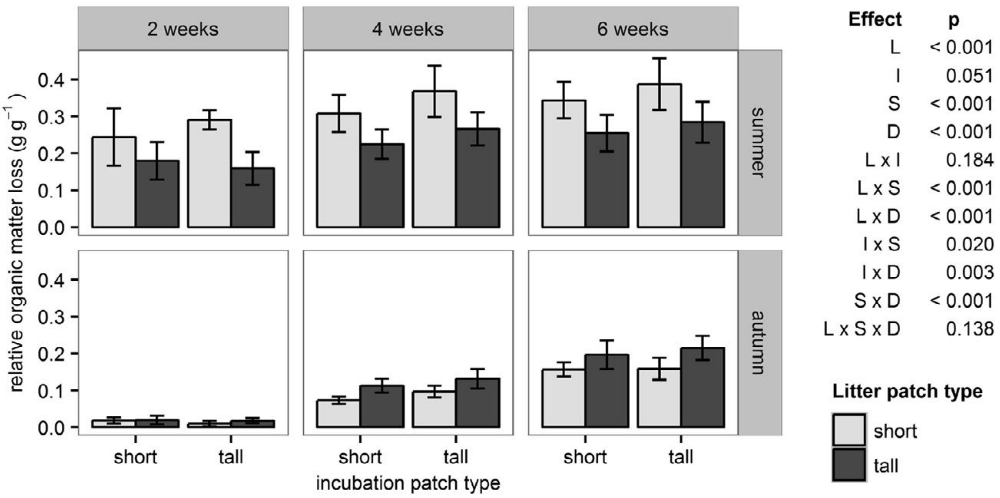

Figure 1. Relative organic matter loss of litter from short or tall patches incubated 2-6 weeks in either short or tall patches in summer and autumn. Error bars: ± standard deviation. *P*-values of the linear mixed model for the main effects litter patch type (L), incubation patch type (I), season (S) and incubation duration (D), as well as their interactions.

Conclusions

Our results confirm that patch-grazing dynamics in pastures influence not only litter quantity, but also litter quality and decomposition conditions. The effect on decomposition rates, however, seems to both seasonally variable and not fully attributable to the litter C:N ratio as a simple indicator of litter quality. Further study of these processes would be valuable, as they might influence the productivity of vegetation patches in long-term stable sward mosaics. It should, however, be accompanied by an investigation on the spatial distribution of nutrients returned through urine and dung, as a redistribution of nutrients between short and tall patches is another potential influence on nutrient cycling in a patch-grazing system.

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Phytodiversity in nutrient-poor heathlands and grasslands: how important are soil chemical factors?

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Abstract

Central Europe is dominated by intensive farming and the concomitant nutrient input is a main thread to biodiversity in open habitats. The rare case of soils unaffected by agricultural intensification found on a military training area (Grafenwoehr, Bavaria, Germany) was used as an opportunity to study the influence of soil chemical factors on plant species richness in two types of open habitats. Species richness of spermatophytes and soil chemical factors (pH, P, K, Mg) were assessed on a total of 94 relevés situated in heathlands (40 relevés) and grasslands (54 relevés). Averaging over linear mixed models, we showed a distinct decline of plant species richness with decreasing pH in both habitats. Despite generally low soil P content, species richness declined strongly with increasing soil P in grassland. Consequently, to preserve highly diverse open habitats, it is necessary to spare them from anthropogenic nutrient input.

Keywords: open landscapes, plant species richness, nutrients, soil pH, model averaging

Introduction

The outstanding value of open landscapes in terms of biodiversity conservation and ecosystem services is well appreciated (Wilson *et al.,* 2012). However, open landscapes face multiple threats, e.g. agricultural intensification, pollution or land use changes, which have led to a serious decline in these habitats during the last decades (EEA, 2015). In central Europe, almost all areas in which soils are suitable for agriculture are farmed intensively and only the poorest sites are spared. Highly diverse open habitats may persist where military training requirements preclude intensive agriculture (Warren *et al.,* 2007). The present study took advantage of a location that has never been subject to modern agricultural practices. Thus, it was possible to examine the relative importance of soil chemical factors for vascular plant species richness in the absence of anthropogenic enhancement of nutrient availability. The influence of edaphic factors on species richness was compared between sites inherently poor (heathland) and sites more rich in nutrients (grassland).

Materials and methods

The study was conducted on the US Army Garrison Grafenwoehr military training area (GTA) in Bavaria, Germany (49°41'.00' N, 11°46'.00'E). GTA covers ca. 230 km^2 out of which 202 ha and 288 ha are designated as the NATURA-2000 habitat types 4030 European dry heaths (on dystrophic, sandy soils) and 6510 lowland hay meadows (on eutrophic calcareous soils), respectively. Open land management includes mowing of meadows once per year and wildlife grazing, especially by abundant red deer (*Cervus elaphus*). In 2014, vegetation surveys were conducted in 10 heathland and nine grassland sites. At each sampling site, four (heathland) or six (grassland) relevés of 5×5 m size were surveyed, totalling 40 relevés in heathlands and 54 relevés in grasslands. Soil samples of each relevé were analysed for phosphorus (P) via CAL-extraction (Schüller 1969), potassium (K), and magnesium (Mg) content and pH-value (pH). Statistics were performed by R statistical software (v 3.2.2; R Core Team 2015). To analyse whether soil chemical factors and their pairwise interactions had a significant influence on species richness, we calculated separate linear mixed effects models (LME) for each habitat type. Sampling site was included as random factor. Data were centralized to zero mean and scaled to 0.5 standard deviation. We compared models using second-order Akaike information criteria (AICc) and performed multi-model averaging in the 'MuMIn' package (Barton 2016).

Results and discussion

On average, we found 14.1±0.8 (mean ± standard error) vascular plant species per 25 m^2 in heathland and 45.9±0.8 species in grassland. The total number of species was 67 in heathland and 154 in grassland. Heathlands and grasslands had distinct species compositions sharing only 20 species. Edaphic conditions generally reflected the lack of fertilisation but differed between habitats. The pH range in heathlands (3.89±0.04) did not overlap with the pH gradient in grasslands (5.72±0.06). P and Mg gradients were lower in heathlands (P: 4.2 0.3 mg kg^{-1}; Mg: 39.2±1.7 mg kg^{-1}) than in grasslands (P: 11.7±7 mg kg^{-1}, Mg: 139.1±11.4 mg kg^{-1}). K availability was similar in heathlands (47.3±2.7 mg kg^{-1}) and grasslands (52.1±1.8 mg kg^{-1}).

Averaging over all LME models explaining the number of plant species per relevé based on soil chemical factors, we found that in heathland, pH, and in grassland, pH and P were the most important variables exerting a significant effect (P<0.05) on species richness (Table 1). In grassland, various additional soil chemical factors and their interactions were present in the set of best fitting models, while in heathland, the best models contained only single fixed effects of pH, K, and Mg. Elevated pH was positively related to species richness in both habitats, most markedly in heathland (Figures 1A and 1B). Even if pH does not have a limiting effect on plants *per se*, it alters nutrient availability (Roem and Berendse, 2000) and affects element toxicity (Roem *et al.,* 2002). This could explain why the negative association between pH and species richness was more pronounced under acidic than under rather neutral conditions. Grassland species richness strongly decreased with increasing extractable soil P content (Figure 1C). The adverse effect of enhanced P availability on grassland diversity has been established by numerous studies including managed and semi-natural areas (Ceulemans *et al.,* 2014; Janssens *et al.,* 1998). The present results highlight that the negative relationship between P and grassland species richness persists even when soil P content is very low.

Table 1. Summary of the linear mixed effects model averaging to analyse vascular plant species richness in heathlands and grasslands in relation to soil chemical factors. Each variable's relative importance represents the sum of Akaike information criteria (AIC) weights within the best candidate models (ΔAIC<2). The estimate is averaged over all best candidate models containing the explanatory variable.

Habitat	Variable	Relative importance (%)	Averaged estimate	SE	z	*P*-value
Heathland	pH	100	7.79	1.37	5.46	<0.001
	Mg	30	0.45	0.96	0.46	0.644
	K	20	0.23	0.71	0.32	0.751
Grassland	P	100	-6.86	2.09	3.21	0.001
	pH	100	3.97	1.62	2.38	0.017
	K	60	1.43	1.65	0.85	0.393
	Mg	45	-1.07	2.09	0.50	0.615
	K×Mg	45	2.26	4.15	0.54	0.589
	Mg×P	28	-4.61	6.02	0.76	0.448
	P×pH	18	-0.46	1.41	0.32	0.751
	Mg×pH	9	-0.39	1.62	0.24	0.813

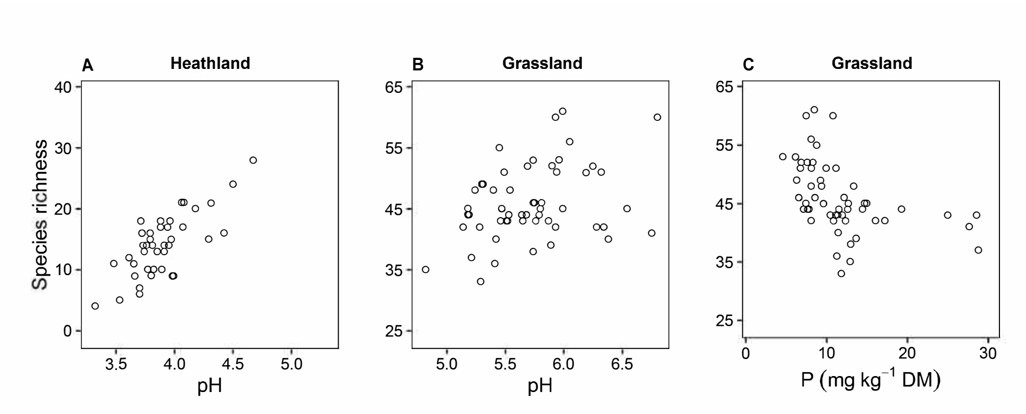

Figure 1. Correlations between vascular plant species richness and pH in (A) heathland and (B) grassland and (C) between plant species richness and soil P content in grassland.

Conclusions

Compared to heathlands, grasslands seemed to be more complex systems, because models explaining plant species richness contained several interacting edaphic factors. Hence, when nutrient availability is elevated, more soil chemical factors may play a role in determining plant species richness than under extremely oligotrophic conditions. With decreasing soil pH, the negative correlation between pH and plant species richness becomes more distinct, indicating toxicity effects. The decrease of plant species richness with decreasing pH and increasing soil P content emphasizes the compelling need to preserve open habitats of high nature conservation value from atmospheric pollution and fertilisation.

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Attractiveness of main sward types to suckler cows grazing on heterogeneous fen grassland

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Abstract

Extensive grazing dominates the agricultural use of formerly drained peat soils in Northeast Germany. Surface profile, and hence sward composition of grazed fen grassland, become more and more heterogeneous, resulting in a patchy vegetation pattern. Detailed knowledge about forage acceptance of the main vegetation types of grazing cattle is crucial to optimize pasture management. A grazing experiment was therefore implemented to evaluate the attractiveness of the main vegetation types for grazing cattle. Two herds of suckler cows were kept on different paddocks of a heterogeneous pastures site: the sward composition in the paddocks at the moderate moist pasture site was almost homogeneous, that of the moist pasture site very heterogeneous. Different sward types were identified and mapped by a GPS-mapper. The leader cow of the herd was equipped with a GPS-collar, recording the movement within the pastures in five minute intervals throughout the grazing season. The surface profile was derived from a digital elevation model. The comparison of the herd's locations and the mapped surface profile reveal minor influence of the altitude on the preferred herd positions during grazing. In the wet heterogeneous pastures the herd spent more than 60% of the grazing time on a sward dominated by species with higher forage value.

Keywords: selective grazing, fen grassland, forage attraction, GPS-tracking

Introduction

Drained fen soils in Northeast Germany are mainly used as grassland (Luthardt and Zeitz, 2013). Extensive grazing by suckler cows is the dominating use of such sites if lower groundwater tables and consequently rather wet soil conditions occur. As a consequence of the long term drainage and degradation of the upper peat horizons the surface profile, the water levels and hence the botanical composition of fen grasslands become more and more heterogeneous. The vegetation adapted to the wet parts is characterized by nutrient-poor species, i.e. *Juncus articulatus*. In opposite, ruderal species and weeds like *Elymus repens* and *Holcus lanatus* become predominant on the higher located areas of the fens (Leipnitz and Fischer, 1997) if no re-seeding measures are applied. These different vegetation patterns result in different attractiveness for grazing cattle and hence in overgrazing or avoidance of the local forage resources (Mladek *et al.,* 2013). Therefore, detailed knowledge about forage acceptance of the main vegetation types is crucial to optimize pasture use efficiency for grazing cattle. A grazing experiment with suckler cow herds was conducted to evaluate the attractiveness of the main vegetation types for grazing cattle over a grazing season.

Materials and methods

The study was carried out on 6 paddocks of a fen pasture at Paulinenaue, Northeast Germany (latitude 52°68'N, longitude 12°72'E, mean annual temperature 9.2 °C, mean annual precipitation 534 mm) in 2014. The surface profile of the experimental site was derived from a digital elevation model DGM 2 (LBGR, 2014). It varied within the pasture area considerably resulting in unequal groundwater-surface distances and hence parts with rather wet, moist or moderate moist conditions.

The paddocks 1 to 3 were located in the moderately moist part of the pasture area. Due to re-seeding in 2013, the swards of these three paddocks were homogenised and consisted of high yielding grasses (Table 1). In contrast, the paddocks 4 to 6 were located in the moist part of the pasture area. Within each of the paddocks 4 to 6 parts with wet, moist and moderately moist soil conditions occurred, resulting in different sward types. The borderlines between those sward types were mapped by a GPS-mapper. The average paddock size was 1.14 (0.95-1.39) ha. For the description of forage quality sward samples were analysed by NIRS technique and also described by the land utilization indicator values (Briemle *et al.,* 2002; Klotz *et al.,* 2002) where the span of values ranges from 1 (least appropriate) to 9 (most appropriate).

The paddocks were grazed in a rotational system by two suckler cow herds with each six cows and one bull at an average stocking density of 6.3 Livestock Units ha^{-1} in 2014. The leader cows were equipped with a GPS collar (Vectronic Aerospace Berlin GPS Plus X Collar) recording the movement within the paddocks in a 5 min interval during the grazing period. Due to the rather small size of some of the different sward types within the paddocks 4 to 6, the locations of the herds were analysed and compared between the moderately moist and the moist pasture site on all six paddocks only during the first three grazing days.

Results and discussion

The mapped surface profile of the pasture area varied by about 100 cm, from 28.4 to 29.4 m a.s.l.. There was a significant influence of the altitude on the herd's preferred places (Table 2). Nevertheless, the common interaction between altitude and sward characteristic suggests that it is not altitude-related soil humidity per se which explains the favoured grazing locations, but the humidity-related attraction of the corresponding plant communities.

Table 1. Forage quality data and land utilization indicator (LUI) of different paddock parts (Tukey, $P<0.05$, different letters indicate significant differences).[1]

Pasture	Re-seeded	Old permanent sward, not re-seeded		
		wet	moist	moderate moist
	Lolium perenne (9), *Phleum pratense* (9), *Festuca* spp. (5-9)	*Juncus articulatus* (2), *Carex hirta* (3), *Alopecurus geniculatus* (5)	*Phalaris arundinacea* (6), *Poa trivialis* (7)	*Elymus repens* (6), *Holcus lanatus* (5)
CP, g kg DM^{-1}	163.1	135.5^{a}	127.0^{a}	116.6^{a}
ADF, g kg DM^{-1}	300.6	334.0^{a}	310.5^{b}	342.8^{a}
ME, MJ kg DM^{-1}	10.18	9.05^{a}	9.72^{b}	9.23^{a}

[1] CP = crude protein; ADF = acid detergent fibre; ME = metabolisable energy; DM = dry matter.

Table 2. Grazing time (hours) at different altitudes of the paddocks (1-6) during the first three days (Tukey-HSD, $P<0.05$, different letters indicate significant differences).

Altitude, m a.s.l.	28.40-28.50	28.50-28.60	28.60-28.70	28.70-28.80	28.80-28.90	28.90-29.00	29.00-29.10	29.10-29.20	29.20-29.40
Mean size, ha	0.120	0.434	0.272	0.192	0.109	0.045	0.028	0.014	0.004
grazing time, h	4.58^{ab}	23.61^{c}	8.89^{b}	5.58^{ab}	3.24^{ab}	0.96^{a}	0.53^{a}	0.31^{a}	0.17^{a}

The grassland sward type had a significant influence on the attractiveness to the suckler cow herds (Table 3). The herd grazing the paddocks 4 to 6 with their old heterogeneous swards spent 60% of the whole day (24 h) on a sward dominated by species with a higher nutritive value. With a share of 17 and 22%, the swards on the moderately moist and the wet parts were more or less avoided by the herd.

Table 3. Grazing time (hours) in different parts of the moist paddocks (4-6) during the first three days (Tukey-HSD, $P<0.05$, different letters indicate significant differences).

Sward type	Area size (ha)	Paddock no.				%
		4	5	6	mean	
Wet	1.16	13.58	14.83	14.92	14.44^a	21.70
Moist	2.36	37.50	41.83	41.75	40.36^b	60.64
Moderate moist	0.64	13.00	11.50	10.75	11.75^a	17.65
Total	4.16	64.08	68.17	67.42	66.55	100

Conclusions

There is a risk of overestimating the attractiveness and hence the feasible contribution of heterogeneous pastures to animal nutrition and liveweight gain. Spatial knowledge on the forage quality of all parts of a paddock is a precondition for a proper pasture management. Depending on the scope of the sward and site conditions, the use of precision farming methods should be assessed in heterogeneous pasture areas.

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Analysis of plant communities growing in small wetland areas in meadows

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Abstract

The aim of the experiment was to assess the role of meadow wetlands in preserving the variety of plants both as individual species and as plant communities. Field research was carried out between 2008 and 2011 in meadow depressions of the Siedlce Plateau in east-central Poland. The area of the reservoirs ranged from 2700 m^2 to 850 m^2. To assess plant communities, homogeneous plots were selected and phytosociological releves were done using the Braun-Blanquet method. The research involved not only plant inventory but also vegetation community analysis together with its classification. Among the plants growing near meadow ponds, are species typical for peat bogs and alder swamp woods but also for fresh and moderately wet meadows or common in bulrush plant areas. The ecosystems studied in the research consisted of a wide variety of rare or endangered species with a considerable diversity in plant communities. The class *Phragmitetea* represented the most numerous group, which is a proof that the wetlands gradually evolve to dry land. The research showed that small water reservoirs in meadows are important in preserving biodiversity among plants and in maintaining plant communities. Moreover, those reservoirs, connecting fragmented ecosystems, are also elements of ecological corridors.

Keywords: flora, plant communities, wetland areas in meadows, east-central Poland

Introduction

One of symptoms of flora synanthropisation is invasion of native species from natural and semi-natural habitats, such and meadow habitats, to distributed habitats. The process does not markedly affect the qualitative changes of flora but it indicates the adaptive attributes that some species have. Due to changes that have recently taken place in the cropping profile, as well as methods and means of agricultural production, agrocenoses are becoming more and more floristically impoverished. At the same time, the compensation of ubiquitous species is increasing, thus destroying structures of segetal communities (Stehlik *et al.*, 2007; Storkey *et al.*, 2011). Native species spreading from meadow habitats increase the floristic diversity of segetal communities (Dąbkowska and Sygulska, 2013; Skrajna *et al.*, 2010).

The aim of the paper is to describe plant communities and the diversity of vegetation in wetland areas in meadows. The research involved not only plant inventory but also vegetation community analysis together with plant taxonomy. The research hypothesis was: natural middle-field aquatic communities situated between intensively used meadows are subject to a process of eutrophication and overgrowing.

Materials and method

The study on plants and plant communities growing in wet depressions in the meadows of the Siedlce Plateau was conducted between 2008 and 2011. The area of the research was located in east-central Poland in the basin of the Bug River. Because of its unique character, over 49% of the Bug River valley is a conservation area protected by law. Six wet meadow depressions were used in the experiment, with an area ranging from 850 m^2 to 27,000 m^2, covering the area of 2.5 thousand km^2. A list of protected species was prepared and homogeneous plots were selected to do phytosociological releves, using the Braun-Blanquet method (Pawłowski, 1977). Tested objects were middle-meadow wastelands, surrounded by intensively used meadows. To carry out the experiment only natural habitats were analysed, with different degrees of tree and shrub coverage. During the four-year research 450 vegetation patches were studied. Classification of plant communities was done using the methods described in Matuszkiewicz (2008) and Matuszkiewicz *et al.* (2012)

Results

There were 312 vascular plant species tested in the experiment. In each experimental unit there were from 126 to 295 species. Synanthropic spontaneophyte species, also called apophytes, together with hemisynanthropic spontaneophytes were the most numerous with kenophytes and efemerophytes being the least common (Table 1). The dominant life form was hemicryptophyte species while ligneous and herbaceous chamephytes were rarely found. The analysis of plant species as parts of plant communities showed that small meadow marshes are mainly overgrown with plants typical for peat bogs and alder swamp woods (*Alnetea glutinosae, Magnocaricion, Scheuchzerio-Caricetea fuscae*) and plants common in wet and medium wet meadows (*Arrhenatheretalia*), but aquatic plants, rushes and water edge shrubs (*Potametea, Semnetea*) are also present there (Table 2).

Table 1. Geographical and historical groups of anthropophytes and origin of apophytes of the vegetation in meadow wetlands.[1]

Research unit	Area	Nr.	Number of species	Sp	Ap	Ar	K	E	M	N	Ch	C	H	G	Hy	T	Nn
	Bujały Mikosze	1	295	128	137	16	8	2	15	20	9	2	144	31	28	45	5
	Łuzki	2	266	88	143	20	11	3	17	16	8	1	131	19	21	53	1
	Zawady	3	220	103	107	5	3	1	13	11	5	2	122	23	23	21	1
	Grądy	4	126	52	73	1	0	0	1	1	3	2	82	11	13	13	0
	Tchórznica	5	223	73	125	16	6	2	15	13	6	1	107	17	21	42	1
	Grodzisk	6	273	114	135	13	8	0	12	15	7	2	144	24	29	40	3

[1] Sp = hemisynanthropic spontaneophytes, Ap = apophytes, Ar = archeophytes, K = kenophytes, E = efemerophytes, M = megafanerophytes, N = nanofanerophytes, Ch = ligneous chamephytes, C = herbaceous chamephytes, H = hemikryptophytes, G = geophytes, H = hydrophytes and helophytes, T = treophytes, Nn = unknown.

Table 2. Sociological groups of the vegetation in meadow wetlands.[1]

Research unit	Area	Nr.	Number of species	Sociological groups																
				1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
	Bujały Mikosze	1	295	30	12	8	4	4	47	36	28	38	25	14	11	9	3	12	6	8
	Łuzki	2	266	28	10	9	3	6	30	29	25	35	25	13	14	10	3	13	8	5
	Zawady	3	220	17	9	4	3	5	46	25	27	38	20	6	6	3	1	4	3	3
	Grądy	4	126	3	2	1	0	0	21	18	21	28	18	8	3	1	0	2	0	0
	Tchórznica	5	223	22	5	7	3	7	24	26	22	33	22	11	12	6	1	11	6	5
	Grodzisk	6	273	19	8	8	4	5	47	33	29	42	25	12	11	6	3	10	6	5

[1] Vegetation groups: 1 = deciduous forests and shrub communities (*Querco-Fagetea, Prunetalia*); 2 = a conifer forest, mixed forest and semi-natural forest (*Vaccinio-Piceetea, Nardetalia, Epilobion*); 3 = nitrophilic shrubs and woodland edge plants (*Sambuco-Salicion, Alliarion*); 4 = xerothermic swards and woodland edge plants (*Festuco-Brometea, Trifolio-Geranietea sanguine*); 5 = psammophilic swards (*Koelerio-Corynephoretea*); 6 = alder wood and peat (*Alnetea glutinosae, Magnocaricion, Scheuchzerio-Caricetea fuscae*); 7 = aquatic plants, bulrushes, wet woods and shrub communities (*Potametea, Lemnetea, Utricularietea, Phragmition, Salicetea purpureae)*; 8 = wet meadows (*Molinietalia*); 9 = fresh and medium-wet meadows (*Arrhenatheretalia*); 10 = flooded nitrophilic swards well-trodden grass (*Plantaginetalia, Agropyro-Rumicion crispi*); 11 = terophytic communities (*Bidentetea tripartiti, Isoeto-Nanojuncetea*); 12 = mesophylic communities of tall perennial plants (*Arction lappae, Convolvulion sepium*); 13 = thermophilic ruderal plants, (*Onopordion acanthii*); 14 = pioneer ruderal communitie (*Sisymbrion officinalis, Eragrostion*); 15 = weed communities of root crops (*Polygono-Chenopodietalia*); 16 = weed communities of cereal crops (*Centauretalia cyani)*; 17 = non-defined plant community.

Altogether 38 plant communities were identified in wet depressions. Plants growing in those depressions were of 7 classes, 9 orders and 16 alliances. The most common community was that of the *Phragmitetea* class. Growing areas of rushes contributes to changing the wet depressions into dry land. The spatial arrangement of plants was mosaic. The biggest floristic and phytosociological diversity was in wetlands flooded with water, intermediate between dry land and wetland.

Conclusions

The vegetation of small wetland areas in meadows is numerous and diversified floristically and phytosociologically (312 species of vascular plants were found, all growing in 38 plant communities). A large share of *Phragmitetea* rushes means that the process of changing into dry land is in progress. Small water reservoirs in meadows are important in preserving biodiversity among plants and in maintaining plant communities. Moreover, these reservoirs, connecting fragmented ecosystems, are also elements of ecological corridors.

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Alternative management for oligotrophic grassland conservation in the Apuseni Mountains

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Abstract

In Eastern Europe, the semi-natural grasslands with extensive management are considered hotspots of biodiversity, some with major international significance. Grasslands with traditional management and high phytodiversity can still be found in vast areas in the Apuseni Mountains (Romania), compared with scattered fragments of extensive grassland in the western countries. Maintaining their multiple functions in the local context, economically feasible solutions are required able to respond also to conservation purposes. Through this study we aimed at assessing the effects of the application of alternative types of management (mowing, mulching, mulching with organic fertilizers and abandonment) on the floristic composition of semi-natural grasslands after 6 years. Our results showed that although organic inputs caused small changes in the floristic composition, without significant changes of the main grassland type. Species like *Festuca rubra* L., *Trifolium repens* L. and others were favoured by mulching and mulching in combination with organic fertilizers (low-input).

Keywords: grassland management, low-input, mulching, organic fertilizers

Introduction

The extensively managed semi-natural grasslands of Eastern Europe (e.g. Romania) are considered biodiversity hotspots of highly international conservation importance (Wilson *et al.*, 2012). Important areas of these grasslands developed under extensive management for many centuries and are now threatened by abandonment, land use changes or intensification. In the Apuseni Mountains the semi-natural grasslands are mostly threatened by abandonment (Păcurar *et al.*, 2015). There are several species and habitats of conservation interest which strongly depend on the continuation of extensive management, particularly mowing (Halada *et al.*, 2011). Researches focusing on the conservation value of the semi-natural grasslands are become more and more frequent, trying to identify alternatives to maintain the extensive management and conserve the biodiversity (Römermann *et al.*, 2009). The aim of our research was to study the effects of mowing, mulching, organic fertilization combined with mulching and abandonment on plant species composition of *Agrostis capillaris* L. – *Festuca rubra* L. grassland, as alternatives to the traditional management, in order to ensure the conservation of semi-natural grasslands in the Apuseni Mountains and prevent abandonment.

Materials and methods

The experiment started in 2009, in Poienile Ursului (1,349 m), Gârda de Sus Commune, Apuseni Mountains, using a randomized block design with 7 treatments in 5 replications: T1 – control (mowed once per year); T2 – mulched once per year; T3 – mulched once per year + 5 Mg ha^{-1} manure (cattle) applied annually; T4 – mulched once per year + 5 Mg ha^{-1} manure every 2 years; T5 – mulched once per year + 10 Mg ha^{-1} manure every 2 years; T6 – mulched once per year + 10 Mg ha^{-1} manure every three years; T7 – abandonment. The floristic composition was described in July 2014 by the Braun-Blanquet method, when the dominant *Poaceae* species were in flower. For the processing of floristic data, we used the PC-ORD, Version 6. NMS (Nonmetric Multidimensional Scaling) was carried out several times in autopilot mode and the process was repeated in manual mode (5 times) in order to minimize the stress (McCune and Grace, 2002). Distance measurement was done with Sorensen (Bray – Curtis). The recommended solution for data presentation was three-dimensional (*stress 15.98*).

Results and discussion

The coefficient of determination *(r^2)* for the correlations between ordination distances and distances in the original *n*-dimensional space was *0.754 (Axis 1 – 0.325; Axis 2 – 0.214; Axis 3 – 0.215)*. Statistically significant experimental factors which explain the changes in floristic composition are: mulching (M), organic fertilization combined with mulching (F). Axis 1 and Axis 2 had no significant correlation with the experimental factors (they are mostly explained by the natural environmental factors), but Axis 3 was positively correlated to M (r=0.472) and F (r=0.368). Significant differences between two factors are expected to be seen in time, as it is shown by Doležal *et al.* (2011), changes in the floristic composition by the low-input treatments are expected after 5 years. Moog *et al.* (2002) have demonstrated that the decomposition of organic matter in mountain areas could take several growing seasons and the effects of mulching could be amplified by litter accumulation over years. Even if the extensive management has no visible effects on vegetation after 10 years (Pavlů *et al.,* 2011), the changes could be evident in soil (Kvítek *et al.*, 2001). Even if most of the species presented weak reactions to the treatment, statistically significant changes were registered for several species (Table 1). For instance, mulching (M) and mulching associated with organic fertilizing (F) had a positive effect (r=0.46) on *Trifolium repens* L. Positive effects of mulching on *T. repens* were highlighted by Zelený *et al.* (2001), the effects being positive in some cases even on the whole group of the *Fabaceae* beside herbs (Mašková *et al.,* 2009). In the studies of Gaisler *et al.* (2004) the share of *T. repens* L. increased from 1 to 21%. Species with weak correlation were: *Anthyllis vulneraria* L., *Lotus corniculatus* L., *Trifolium pratense* L., *Achillea distans* Waldst. et Kit. ex Willd., *Alchemilla vulgaris* L., *Arnica montana* L., *Centaurea mollis* Waldst. et Kit., *Colchicum autumnale* L., *Leucanthemum vulgare* Lam., *Pimpinella major* (L.) Huds., *Polygala vulgaris* L., *Primula veris* L., *Ranunculus acris* L., *Rhinanthus minor* L., *Trollius europaeus* L. and *Veratrum album* L.

Conclusions

Four of the treatments caused no changes in the floristic composition after six years, not even that with abandonment. Mulching in association with organic fertilization changed (increased/decreased) the share of some of the species. The treatments could be taken into consideration for conservation purposes, considering their minor effects upon the floristic composition after 6 years.

Table 1. Correlation of species (significant) with the ordination Axis 3.

Species	Axis 3	
	correlation coefficient (r)	Significance[1]
Festuca rubra L.	0.382	*
Trifolium repens L.	0.466	**
Astrantia major L.	-0.453	**
Hieracium aurantiacum L.	0.457	**
Hypericum maculatum Crantz	-0.512	**
Linum catharticum L.	-0.412	*
Plantago lanceolata L.	-0.508	**
Polygala comosa Schkuhr	-0.406	*
Ranunculus bulbosus L.	-0.378	*
Thymus pulegioides L.	0.387	*
Veronica chamaedrys L.	-0.541	***

[1] * P<0.05, ** P<0.01, *** P<0.001.

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The effects of mulching and mineral fertilizers on oligotrophic grasslands' floristic composition

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Abstract

Mulching of grasslands has been suggested by several researchers as a feasible solution to maintain the natural productivity and floristic diversity of low productive systems when the succession of woody species is a threatening factor. In this mid-term experiment (4-5 years), we aimed to study the effects of new management alternatives in the Apuseni Mountains (Romania), such as mulching and mulching combined with mineral fertilization (NPK, PK), on the floristic structure of oligotrophic grasslands. After five years, there were no significant changes to the main grassland type, but significant changes had taken place in the floristic compositions of the experimental plots. The overall effects of the treatments were negative upon the phytodiversity in some of the variants. For this reason these should be recommended with caution in the conservation of oligotrophic grasslands.

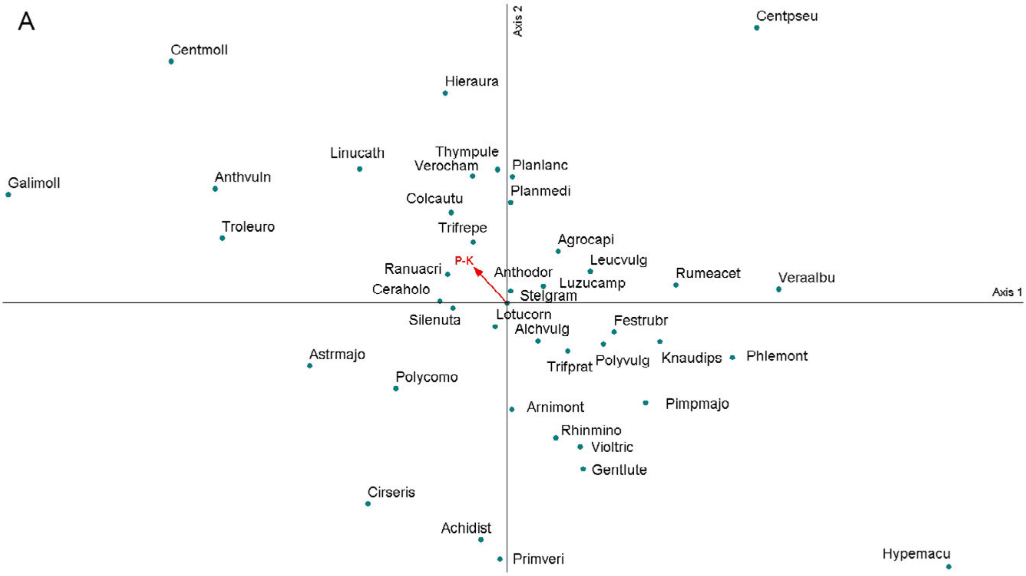
Keywords: mulching, mineral fertilizers, oligotrophic grassland, biodiversity

Introduction

Oligotrophic grasslands are important from the biodiversity point of view. The traditional grassland management (summer hay making followed by autumn grazing) in mountain areas is no longer economic, thus alternative solutions are required (Doležal *et al.*, 2011). In the last decade, in many studies, mulching was mentioned as an alternative option to maintain minimum disturbance and to assure low nutrient input for grassland ecosystems. Gaisler (2006) and Mašková (2009) suggested that mulching could be a feasible solution for the low productive systems to maintain their natural productivity and floristic diversity, when the succession of woody species is a threatening factor (Carboni *et al.*, 2014). Tonn *et al.* (2010) highlighted that mulching could be an alternative solution also for the conservation of semi-natural grasslands with high level of biodiversity. Mineral fertilizers are forbidden for conservation purposes in compensatory payment schemes in many of the EU countries. However, it could be that farmers will use mineral fertilizers on oligotrophic grasslands, despite low economic feasibility. Following the hypothesis that traditionally managed oligotrophic grasslands are in functional equilibrium, the question is which management treatment will maintain communities whose floristic composition is most similar to that of the traditional management regime. The aim of this study was to identify the suitability after five years of different management types using mulching and mulching combined with mineral fertilizing on *Agrostis capillaris* L. – *Festuca rubra* L. grasslands in the Apuseni Mountains.

Materials and methods

The experiment was carried out in 2009, in Poienile Ursului (1349 m.a.s.l.), Gârda de Sus Commune, Apuseni Mountains, in randomized block design with 7 treatments in 5 replications: T1 – control (mowed year^{-1}); T2 – mulching year^{-1}; T3 – mulching 2× year^{-1}; T4 – mulching year^{-1} + N25:P25:K25 applied annually; T5 – mulching year^{-1} + 1× in 2 years N25:P25:K25; T6 – mulching year^{-1} + P25:K25 applied annually; T7 – mulching year^{-1} + 1× in 2 years P25:K25. The floristic composition was interpreted using the Braun-Blanquet method, when the *Poaceae* species were in flower (in July, 2013). For the processing of floristic data, we used the PC-ORD, Version 6. NMDS (Nonmetric Multidimentional Scaling) was carried out several times in autopilot mode and the process was repeated in manual mode (5 times) in



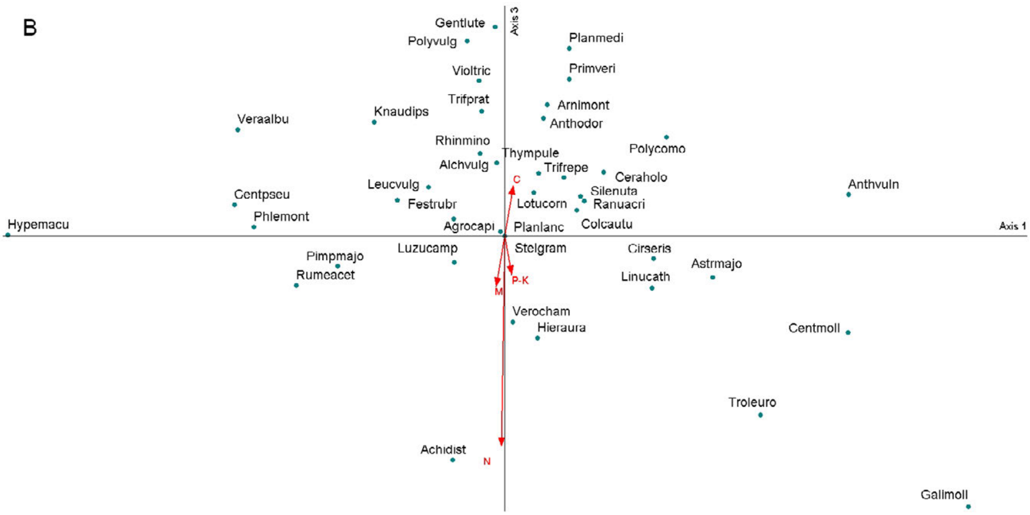


Figure 1. (A) Influence of the treatments on plant species composition (Axis 1 and Axis 2). (B) Influence of the treatments on plant species composition (Axis 1 and Axis 3). A = Abandonment, C = Mowing, M= Mulching, N = Mulching with mineral fertilizing N25:P25:K25, P=K = Mulching with mineral fertilizing P25:K25, Agrocapi = *Agrostis capillaris* L., Alchvulg = *Alchemilla vulgaris* L.; Arnimont = *Arnica montana* L.; Centmoll = *Centaurea mollis* Waldst. et Kit.; Centpseu = *Centaurea pseudophrygia* C.A.Mey.; Ceraholo = *Cerastium holosteoides* Fr.; Cirseris = *Cirsium erisithales* (Jacq.) Scop.; Colcautu = *Colchicum autumnale* L.; Euphoffi = *Euphrasia officinalis* L.; Festrubr – *Festuca rubra* L., Galimoll = *Galium mollugo* L.; Gentlute = *Gentianella lutescens* (Velen.) Holub; Hieraura = *Hieracium aurantiacum* L.; Hypemacu = *Hypericum maculatum* Crantz; Knaudips = *Knautia dipsacifolia* Kreutzer; Leucvulg = *Leucanthemum vulgare* Lam.; Linucath = *Linum catharticum* L.; Lotucorn = *Lotus corniculatus* L.; Pimpmajo = *Pimpinella major* (L.) Huds.; Planlanc = *Plantago lanceolata* L.; Planmedi = *Plantago media* L.; Polycomo = *Polygala comosa* Schkuhr; Polyvulg = *Polygala vulgaris* L.; Primveri = *Primula veris* L.; Ranuacri = *Ranunculus acris* L.; Rhinmino = *Rhinanthus minor* L.; Rumeacet = *Rumex acetosa* L.; Silenuta = *Silene nutans* L.; Stelgram = *Stellaria graminea* L.; Thympule = *Thymus pulegioides* L.; Trifprat = *Trifolium pratense* L.; Trifrepe = *Trifolium repens* L.; Troleuro = *Trollius europaeus* L.; Veraalbu = *Veratrum album* L., Verocham = *Veronica chamaedrys* L.; Violtric = *Viola tricolor* L.

order to minimize the stress (McCune and Grace, 2011). Distance measurement was done with Sorensen (Bray – Curtis). The recommended solution for data presentation was three-dimensional (*stress 12.22*). The coefficient of determination *(r^2)* for the correlations between ordination distances and distances in the original *n*-dimensional space was *0.864 (Axis1 – 0. 489; Axis2 – 0.243; Axis3 – 0.132)*. NMDS applied on the plot level did not show any significant results. We therefore chose not to present those results in this paper.

Results and discussion

Small changes occured in vegetation determined by the treatments, which took place within the *Agrostis capillaris* L. – *Festuca rubra* L. grassland type. Vectors like mowing (C) and mulching (M) and fertilizing with P25:K25 (P-K) are not significantly correlated with any of the axes. Fertilizing with N25:P25:K25 (N) is negatively correlated with Axis 3 (*r*=-0.540).

There are few studies, both international or national, that have looked into the effects of mulching combined with mineral fertilizing. In Romania, Păcurar *et al.* (2011) investigated the effects of mulching on oligotrophic grasslands in the Apuseni Mountains. The results of Bogdan (2012) demonstrated that mulching in combination with mineral fertilizers causes no changes in the phytodiversity in the short term (1-3 years). As presented in Figure 1, fertilizing with N25:P25:K25 (N) had a positive influence on species like *Achillea distans* Waldst. & Kit. ex Willd., *Galium mollugo* L. and *Trollius europaeus* L. However, the same fertilizer treatment had a negative effect on the spices *Arnica montana* L., *Plantago media* L., *Anthoxanthum odoratum* L., *Gentianella lutescens* (Velen.) Holub, *Knautia dipsacifolia* Kreutzer, *Polygala vulgaris* L., *Viola tricolor* L. Negative effects of mineral fertilizers in higher doses upon *A. montana* L. and *P. media* L. were mentioned also by Brinkmann *et al.* (2007).

Conclusions

The experimental treatments caused no major changes in the floristic composition and the grassland was classified as the same vegetation type even after five years. Thus, mulching (without or in combination with fertilizing) can be considered as an alternative (e.g. to abandonment or intensive use) for maintaining the floristic composition of oligotrophic grasslands in the short term. However, whether this treatment can be recommended on a large scale should be carefully analysed in relation to the biodiversity conservation goals. The long-term effects upon certain species (e.g. *A. montana*) should also be considered.

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Grassland: quantification of the environmental services provision

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Abstract

Grasslands provide a multitude of environmental services. These services include the provision of forage and habitats for species-rich communities as well as carbon storage, erosion protection and flood retention. Unfortunately, the services are not always synergetic. In particular farmers can often generate more forage or higher income if they intensify grassland management or convert it to arable land. This comes frequently at the expense of a lower provision of other services. We use geographic information systems (GIS) in order to quantify the role of grassland for the provision of different services and to identify hot spots for the conservation and reestablishment of grassland in Germany. The results show that more than 56% of grassland in Germany is important for the provision of environmental services. Nearly 30% of the German grassland is on slopes steeper than 8% and contribute to the prevention of soil erosion. More than 20% has a high importance for preservation of biodiversity and 15% of the German grassland is located on carbon-rich soils. In particular in large parts of North Eastern and Central Germany, the overwhelming share of the remaining grassland is important for the provision of environmental services.

Keywords: grassland, GIS-analysis, environmental services

Introduction

Grasslands provide a multitude of environmental services. These services include the provision of forage and habitats for species-rich communities as well as carbon storage, erosion protection and flood retention. In Germany the conservation status of most grassland habitat types listed in the Habitats directive (EEC 92/43) is bad or insufficient and the population of many species typical for meadows and pastures has declined dramatically within the recent years (e.g. BfN, 2014). On organic soils the greenhouse gas emissions of grasslands are positively correlated to the land use intensity and the level of drainage, and the emissions of intensively used grassland are of the same magnitude as arable land (e.g. Drösler *et al.*, 2014). The year-round vegetation cover of grassland reduces the erosion risk and is beneficial with respect to flood retention. Another aspect is the protection of water quality, as nitrate leaching below grasslands is significantly lower compared to arable land (BMUB, 2013). However, aspects of water protection are not addressed in this analysis. For all these reasons the protection of grassland and their adequate management is an important issue in the German agri-environmental debate. However, between 1995 and 2015 the extent of grassland declined in Germany by 11% while the extent of arable land remained stable (Destatis, various years). Many farmers convert grassland to arable land because arable land is more profitable. A quantification of the grassland area is important in order to quantify environmental services. This paper intends to bridge this gap.

Materials and methods

Our analysis is based on various data sources that are available at the national level. The data are processed on a 100 m grid. Table 1 provides an overview of the used databases and a brief description. We conducted a geostatistical analysis using a suite of software packages (PostgreSQL 8.4.5, ARCGIS 10, PostGIS 1.5.2 and SAS 9.3) for the processing and statistical analysis. Laggner *et al.* (2014) provides details on the data, the work flow and the processing.

Table 1. Data sources used in this study.

Dataset	Source	Year	Purpose
Digital Landscape Model	BKG	2010	Delimitation of grassland
Digital Terrain Model	BKG	2009	Slope
Geological map	BGR	2003	Distribution of organic soils
Areas with conservation status	BfN	2010/2011	Areas with conservation status
Designated Floodplains	Various federal states	2002-2012	Distribution of designated floodplains

For this analysis we focus on three environmental services: erosion prevention, biodiversity preservation and climate mitigation. We define grassland as being important for the provision of environmental services if at least one of the following conditions is fulfilled: the grassland is located in either a designated floodplain or on slopes that are steeper than 8% (erosion prevention), in a Special Area of Conservation (Habitats Directive), in a Special Protection Area (Birds Directive EEC 79/409) or in a nature conservation area (biodiversity), or on organic soils (climate mitigation).

Results and discussion

In 2010 4.6 million ha or 28% of the utilized agricultural area in Germany was used as grassland. The pre-alpine area and the areas along the Wadden Sea are the only areas where grassland is the main component of the landscape. In the uplands the open landscape is mainly used as grassland, whilst forests are here the main land cover. 56% of the German grassland has high importance for the provision of the analyzed environmental services. In most regions over 75% of the grassland is important for the provision of environmental services (Figure 1). Low shares can be found in the pre-alpine belt near the Alps, in North-Eastern Germany and along the North Sea. Here grassland is frequently intensively used by dairy farms. In many areas with a low share of grassland, e.g. North-Eastern Germany and southern parts of Lower Saxony, the grassland is concentrated in designated areas or areas with specific site conditions, e.g. flood plains or organic soils.

Over 20% of the German grassland is located in areas designated for the protection of biodiversity and 14% on organic soils. As these categories are spatially overlapping, on 29% of the German grassland an intensive grassland use has significant trade-offs with respect to climate mitigation and the protection of biodiversity. Of the remaining grassland with an importance for the provision of environmental services, the overwhelming majority contributes to erosion protection (24% of the total). 3% of the grassland is located on floodplains outside designated areas and not on organic soils. The maintenance of the entire grassland area avoids greenhouse gas emissions from conversion into arable land. Thus, for climate mitigation 100% of the grassland area is important. However, on most of the area, the intensity of grassland use is not relevant for this aspect.

The analysis shows that the potential for the intensification of grassland is limited if the environmental services provided are not to be set at stake. Also, intensive use is frequently limited due to the site conditions (soil moisture, inclination). With respect to an increased provision of environmental services from agricultural land, the issue must be addressed whether an optimization of the distribution of arable land and grassland can be achieved with the help of the prior authorization scheme of the Greening. In Germany farmers can only convert grassland into arable land if an arable area of the same size is converted to grassland, and only with the consent of water and nature conservation authorities. An option to improve the environmental service provision is to allow the conversion only if the area does not comply with any of the above mentioned criteria. This option is implemented by two federal states in their regulatory law (Schleswig-Holstein and Mecklenburg-Vorpommern).

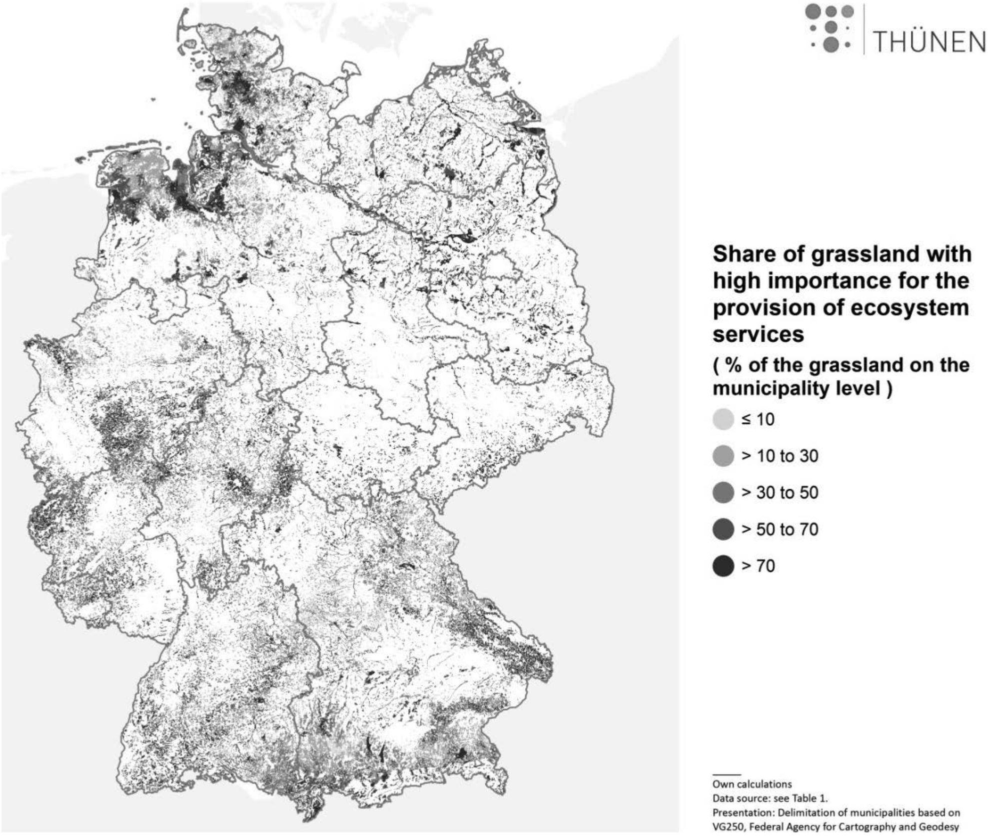

Figure 1. Distribution of grassland with a high importance for the provision of environmental services (biodiversity, carbon storage on organic soils, erosion protection and flood retention).

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Leaf temperature as a proxy for competition in phytodiverse agricultural grassland

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Abstract

Phytodiverse grassland has been suggested to use resources like water, light and nutrients in a more efficient way than mono-species stands as the (intraspecific) competition for these resources should be smaller in diverse swards. However, it is difficult to measure competition for resources directly in established grassland swards. The temperature of the youngest leaves of grassland plants may potentially serve as a proxy for competition for water: plants with long roots that have more water available can cool down as an effect of transpiration. More available nutrients, especially nitrogen, can lead to more biomass production per unit of water used, with varying effects on leaf temperatures: a decrease due to water savings or an increase due to taller heights. Tall plants successfully competing for light may have higher leaf temperatures due to increased interception of the sun's radiation, but this depends on the arrangement and position of leaves. Here, we measured temperatures of the youngest leaf of grasses and forbs in fertilized and unfertilized phytodiverse grassland to test whether leaf temperature may serve as a proxy for competition in these settings.

Keywords: biodiversity, water, infrared, *Dactylis glomerata*, *Rumex acetosa*, *Taraxacum officinale*

Introduction

Phytodiversity has become a major point of interest for agricultural and ecological scientists. In previous studies, plant diversity has been suggested to be important for productivity in grassland ecosystems, probably resulting from an improved complementarity in resource use (Naeem *et al.,* 1994, Tilman *et al.,* 1996). However, many studies of the biodiversity – productivity relationship have been carried out on experimental grassland, leading to results not readily applicable to permanent agricultural grassland (Caldeira *et al.,* 2001; Isselstein, 2005). Furthermore, the determination of (inter- and intraspecific) competition of plants for resources is often cost- and labour-intensive (e.g. determining competition for water by indicators like carbon isotope ratios). Being able to investigate the competition for water among plant species via leaf temperature measurements, as a proxy for a plant's transpiration, would greatly simplify the research work. Hence, we carried out a first experiment in established phytodiverse grassland, to test whether the temperature of the youngest, well-developed and healthy leaf of the tested plants differs. If so, this may potentially serve as a proxy for competition among plant species; fertilisation (expected to increase competition), different rooting depths and plant heights may modify this relation.

Materials and methods

The experiment was set up on permanent grassland, an oat grass meadow on loamy sand, at the experimental station of the University of Rostock, Germany (54°3'39' N, 12°4'52' E). The grassland is managed extensively by cutting, with no fertilisation. For the experiment, ten plots of 1 m^2 each were established, five of which were fertilised with calcium ammonium nitrate on 7 May and again after a cut (height 8 cm) on 27 May 2015 (300 kg ha^{-1} per application, or 81 kg N ha^{-1}). The species chosen for leaf temperature measurements based on their occurrence and their relatively large leaves were *Dactylis glomerata*, *Rumex acetosa* and *Taraxacum officinale*. The infrared handheld thermometer Optris MS was used to measure the temperature of the youngest, fully developed leaf per plant. Per plot, one plant of each target species was measured three times and data averaged per species and plot. Measurements were done on 15 occasions ('day') with little wind and stable conditions, between 20 May and 15 July 2015. The data was analysed with R (version 0.99.491) using linear mixed effects models with 'treatment' (fertilisation yes/no) and 'species' as fixed effects and 'plot' and 'day' of the experiment as random effects (R Core Team, 2014). Differences were analysed with ANOVA and the Tukey-test (α=0.05). Literature data on rooting depth and plant height were used to discuss potential influences on leaf temperature (Kutschera *et al.,* 1982, 1992).

Results

Fertilisation did not significantly influence plant temperatures (P>0.599*).* Overall, the temperatures of the unfertilised plots were slightly higher than those of fertilised ones (23.1 versus 22.8 °C for unfertilised and fertilised, respectively). In contrast, the factor species had a significant effect on temperatures (P<0.001; Table 1). Over all measurement days, the temperatures of *R. acetosa* and *T. officinale* were significantly higher than those of *D. glomerata.* In seven of the fifteen measurements, the factor species was significant. In five of these seven measurements, *R. acetosa* had the highest measured temperatures (run 4 to 8). *T. officinale* had the highest temperatures in the two remaining measurements showing significant differences (run 11 and 14). There was a clear negative correlation between plant height (from literature data) and temperature. The tallest plant (*D. glomerata*) was at the same time the plant with the lowest temperature measurements. *R. acetosa* was the smallest plant with usually the highest temperatures. For the root depth, we could not find a correlation with temperature. Day of the experiment as a random effect explained about 21% of the variance in the model, compared to the fixed effects that explained only about 3%. The random effect plot had a low rate of explained variance of about 0.5%

Discussion

Unexpectedly, fertilisation had no significant effect on plant temperature in this short-term experiment. However, the trend of the data showed lower temperatures with the use of fertilisation. Shading due to better growth in fertilised plots should not have affected measured temperature as we always measured the youngest (unshaded) leaf. Thus, the temperature trend may have been due to better water availability and thus more transpirational cooling in fertilised plots. However, a larger distance from the ground of better-growing plants could also have influenced temperatures, as discussed below. Differences in temperatures among plant species were observed especially after the cut that took place before measurement day 4. After this, species with remaining leaves close to the ground (*R. acetosa* and *T. officinale*) had higher temperatures until the biomass regrew (measurement day 4 to 8, Table 1). This indicates that solar radiation reaching the ground and the leaves directly had a large effect on leaf temperatures. The more upright architecture of *D. glomerata* probably made this species less prone to direct heating by the sun and allowed better cooling by wind. Thus, plant height seems to have had a larger influence on plant temperature, also due to warming of the ground in this relatively open stand, than the rooting depth, which was expected to be smallest in *D. glomerata*. The generally small explained variance indicated that the effect of more factors on leaf temperature have to be considered (e.g. soil temperature, wind speed and transpiration), in order to be able to use leaf temperatures as proxies for competitive abilities of different plant species in permanent grassland. To improve measurement precision, measurements of all plants should be made at a similar height above the ground. The use of IR cameras could help to get a better integrated measurement of temperatures per plant, thus decreasing the variation in the data. As differences among plants have been shown to be small, the device used needs to be very sensitive.

Conclusions

Temperature measurements of plant leaves may help understand competitive behaviour of different plant species in mixtures. However, effects of the distance of measured leaves from the ground need to be considered. Sensitive measurement techniques are needed to identify the small differences among plants in permanent grassland.

Table 1. Average temperature values (°C) of the youngest leaves of *Dactylis glomerata* (*Dg*), *Taraxacum officinale* (*To)* and *Rumex acetosa* (*Ra*) for each measurement occasion (day), with standard deviation in brackets.[1]

Day	Dg	To	Ra
1	16.5 (3.3)	18.6 (4.0)	16.4 (2.8)
2	13.1 (2.8)	12.5 (3.5)	12.4 (1.9)
3	18.6 (1.0)	19.0 (0.7)	18.8 (0.4)
4	21.6 (1.5)[a]	24.3 (3.3)[b]	27.0 (3.3)[c]
5	22.3 (4.1)[a]	24.7 (1.3)[ab]	26.6 (2.4)[b]
6	21.7 (4.2)[a]	25.8 (3.2)[b]	27.7 (3.0)[b]
7	22.0 (3.1)[a]	24.7 (2.5)[ab]	26.5 (4.7)[b]
8	28.2 (4.8)[b]	27.9 (1.2)[a]	32.0 (3.5)[b]
9	20.2 (2.7)[ab]	19.5 (1.5)[a]	22.9 (4.5)[b]
10	20.3 (1.5)	19.9 (2.4)	20.6 (2.3)
11	23.5 (1.5)[a]	26.2 (2.3)[b]	24.9 (1.4)[b]
12	26.2 (1.6)	27.2 (2.9)	27.7 (1.4)
13	25.9 (1.3)	27.9 (1.9)	26.6 (3.3)
14	27.4 (1.4)[a]	29.9 (2.4)[b]	28.4 (2.1)[ab]
15	19.9 (3.2)	20.2 (4.4)	20.1 (4.9)

[1] Results of the Tukey test (α=0.05) shown as superscript letters. Different letters indicate significant differences among species. Where no letters are shown, there were no significant differences.

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The effect of soil P on legume distribution and biodiversity on small spatial scales

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Abstract

N fixing legumes play an important role in providing N and achieving high forage quality, especially in organically managed grasslands. Many farmers observe both temporal and spatial variation of the legume distribution on their grassland sites. Legumes have high P requirements, so that legume and total yield can be increased by P fertilisation. However, large concentrations of soil P negatively impact phytodiversity. In our study we investigated whether the concentration of soil P explains the presence of legumes and if so, which P fraction is most appropriate for this. Additionally, we examined whether an effect of soil P on biodiversity could be ascertained. The study was conducted on two mown pasture sites under organic management in north-eastern Germany. Here, several plots were investigated with a clear spatial differentiation between the functional groups 'legumes' and 'non-legumes'. The species richness of each plot was recorded. Vegetation and soil samples were taken and analysed separately for legumes and non-legumes. Soil P contents of the different fractions and thus P availability could not explain the spatial distribution of legumes and no relation between soil P and biodiversity was found.

Keywords: *Trifolium repens*, *Trifolium pratense*, *Lotus corniculatus*, *Lathyrus pratensis*, Hedley phosphorus fractionation

Introduction

In semi-natural grasslands, productivity often conflicts with biodiversity. P in soil is heterogeneously distributed and differs in availability. It may affect both phytodiversity and vitality of legumes, thus influencing productivity (Ceulemans *et al.*, 2014; Høgh-Jensen *et al.,* 2002). The objective of this study was to investigate if there is a relation between (1) soil P fractions and the presence of legumes and (2) between soil P and biodiversity.

Materials and methods

The study was conducted on two mown pastures (*Lolio-Cynosuretum*) on sandy mineral soil under organic management in north-eastern Germany. In September 2015 we investigated twelve plots of 2×2 m showing a clear spatial differentiation between the functional groups 'non-legumes' and 'legumes'. The 'legumes' were always dominated by either *Trifolium repens* (n=4), *Trifolium pratense* (n=3), *Lotus corniculatus* (n=4) or *Lathyrus pratensis* (n=1). Phytodiversity of each plot was recorded. Separately for each functional group per plot, vegetation and soil samples were taken, dried and analysed using OES-ICP. For vegetation samples of 'non-legumes', only grasses were harvested, no herbs. P concentrations of plant material were analysed after reaction with HNO_3 and H_2O_2 and digestion by microwave. P fractionation of soil samples (0-10 cm) followed the modified Hedley *et al.* (1982) scheme according to Tiessen *et al.* (1983). Additionally, plant available soil P (P_{DL}) was measured using the double lactate method (Riehm, 1948). Statistical analysis was done with R (Version 3.0.2, R Core Development Team, 2011).

Results and discussion

The proportion of legumes in all plots was 12.8±7.8%, of grasses 60.3±10.6% and of herbs 26.8±11.3% (mean±standard deviation). There was a significant difference in plant nutrient contents between 'non-legumes' and 'legumes'(P<0.05, t-test, data not shown), except for K. The P content in 'legumes' was smaller than in 'non-legumes'. Surprisingly, no significant difference could be found concerning their associated soil nutrient contents or the P fractions (P>0.05, t-test, data not shown). This is in contrast to observations by Thomas and Bowman (1998) who found noticeably smaller P contents in clover patches than in surrounding soil. Heuwinkel *et al.* (2004) explained the spatial variability of legume contents of swards with the site-specific availability of soil N, which was not considered here. There was also no significant difference in soil P fractions under the different legume species (P>0.05, ANOVA). Most P was measured in the least extractable NaOH- and H_2SO_4-fractions (Figure 1).

The mean species number of all plots was 13.3±2.4, Shannon index was 1.83±0.35 and Eveness 0.71±0.13 (mean±standard deviation). There was no clear relation between species number and soil P_{DL} content (Figure 2). Ceulemans *et al.* (2014) found a strongly negative relation between plant species number and soil P contents of European grasslands. On average, they found 17.28 species 4 m^{-2} in lowland hay meadows, which is more than we found.

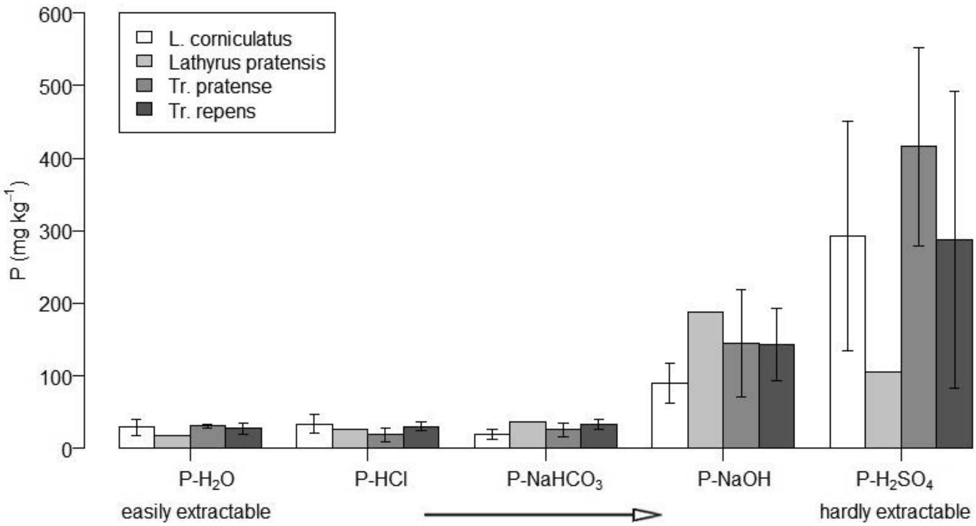

Figure 1. Phosphorus content (mg kg^{-1}) extracted by various solvents from soil samples taken under various legumes species. Results are presented as mean±standard deviation. *Lotus corniculatus* (n=4), *Lathyrus pratensis* (n=1), *Trifolium pratense* (n=3), *Trifolium repens* (n=4).

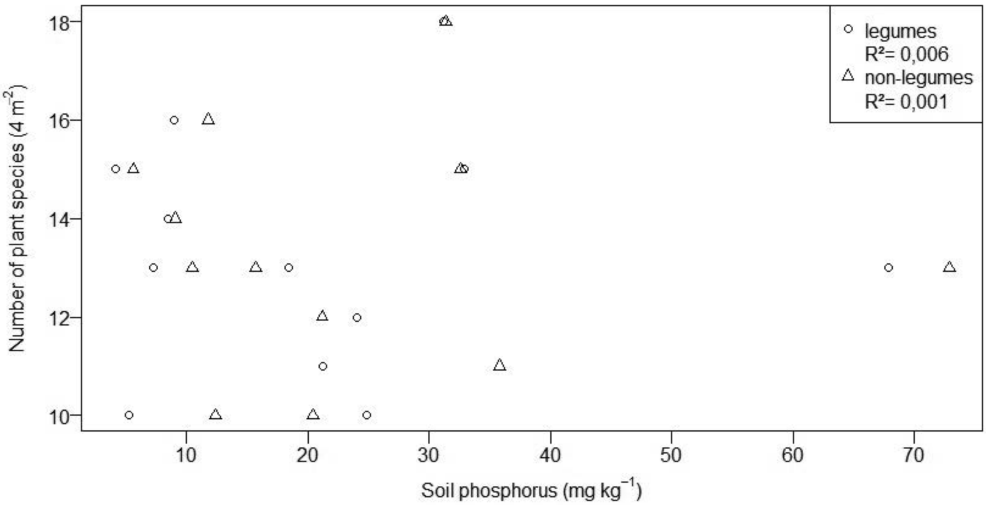

Figure 2. Species number (4 m^{-2} versus soil P content (mg kg^{-1}) measured by double lactate method (P_{DL}). The relation between P_{DL} of 'legumes' and species number of each plot is shown with circles, that of 'non-legumes' with triangles.

Conclusions

Soil P contents of the different fractions and thus P availability could not explain the presence or absence of legumes or the phytodiversity at small spatial scale on an organically managed permanent grassland site.

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Traditional to commercial use of seaweeds: cross-disciplinary perspectives in using local protein sources in Arctic sheep husbandry

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Abstract

To address the increased demand in protein-rich substitute for soya for animal husbandry, knowledge on the potentials for largescale and sustainable cultivation and distribution of local resources containing high proteins is essential. Our project 'Legumes and algae as alternative protein sources in fodder for sheep' (AltPro) is assessing the protein contents and digestibility as well as the social and managerial barriers and opportunities to (re)introduce local protein sources (e.g. seaweeds) in sheep farming. We present qualitative data i.e. interviews with sheep farmers in a coastal community in Northern Norway. The local respondents provide insights on the traditional and present use of seaweeds, and their attitudes toward future large-scale use of these resources. Specific to the northern communities, narrative analyses showed that positive attitudes were associated with the historical and present use of the coastal seaweed resources as feed. Conversely, negative attitudes were associated with the uncertainties related to environmental and ecological impacts, economic margins and the practicalities related to large-scale cultivation and distribution of local protein resources for sheep farming in northern Norway.

Keywords: seaweed, sheep farming, narratives, coastal communities

Introduction

Norwegian meat production is highly dependent on feed concentrate, based on imported soya. An increasing interest for locally produced feedstuff for ruminants is the background for the project 'Legumes and seaweed as alternative protein sources for sheep' (Altpro) that was launched in 2014. The main objective of the project is to identify alternative protein sources, where locally available legumes and seaweeds seem to be the most suitable, sustainable and environmental friendly sources. Use of seaweeds as animal feed is not new; people living on coastal areas have traditionally fed their animals with seaweeds especially during lean feed seasons. Moreover, free-range ruminants had been observed to wilfully graze on beach-cast seaweeds. The use of seaweed as an alternative protein source in animal feed, either as bulk ingredient or as extracted protein component, still requires basic studies (e.g. Tayyab *et al.*, 2016). Moreover, the ecological impacts of using seaweed, either from wild harvest, monoculture or integrated multi-trophic aquaculture (IMTA), and the economics of extracting their protein in a commercial scale also requires further investigation. Alternatively, seaweed may be used as functional food ingredient that can be incorporated into animal feed, i.e. into the pellets, primarily for the various bioactive compounds that may have health benefits. In this paper the historical use of seaweed as animal feed, a practice that had been handed down through generations, and the farmers' perspectives toward large-scale use of seaweeds for animal feed were documented.

Materials and methods

We did a case study of Røst farming community. Røst is located in the outskirt of the Lofoten archipelago in Nordland county (67° North). The municipality consists of more than five hundred islands with rocky outcrops, mountains, lowlands, marshes and shrubs that have been used for pastures throughout the centuries. Earlier the Røst islands hosted more than a thousand people that based their living on combined fishing and farming, the so-called '*fiskarbonden*'. Today approximately 550 people live here. Structural changes in farming have led to a shift toward larger units over the last years (Jørgensrud, 2014) and today only four *fiskarbønder* represent the agricultural sector on Røst, keeping about 600 sheep on the islands. These farmers and two representatives from municipal administration were interviewed. In addition a group interview with six elderly people was made. The respondents were asked to tell their story about how they feed their sheep during the year. In addition, we used a semi-structured interview guide with open-ended questions to get the information we did not receive from their story-telling. Through farm visits, we also got information about the farming facilities, such as animals, barns (sheep shed), fodder applications, grazing fields, as well as of the context of the rural community. Narrative analyses of the qualitative interviews were undertaken to identify main rationales and attitudes related to earlier and present practices related to local feed (Pashcen and Ison, 2014).

Results and discussion

The elderly people we interviewed narrated practices from the 1940s and 1950s when most households kept 8-10 sheep and 1-2 cows. During that time feed concentrate was not available and feed shortage sometimes became a severe problem during late winter. Seaweed was frequently used as alternative and/or supplementary feed all over the coast, and the animals actively seek and feed on beach-cast seaweeds during low tides. Sometimes winter storms cast huge amount of seaweed biomass onshore which make the potential feed easily accessible for the farmers as well as for the animals on pasture to graze. Seaweeds were also brought to the barn during late winter. Multiple practices were used to harvest the seaweed standing stock, either by boat or from the beaches. Knives and scythes were used to cut the seaweeds. The beach-cast seaweeds are usually collected using sled and boxes and stored outside the barn in huge loads. It was labour-demanding and hard work. Most people gave the seaweed directly to the animals, without any processing. Some stories about ways to prepare the seaweed exist, including parboiling.

Figure 1. Sheep feeding on *Palmaria palmata* on the beach at Røst (Photo: Ingrid Bay-Larsen).

Different species of red and brown algae were part of the animal diet. Among the most frequently mentioned species was *Palmaria palmata*. Other seaweeds like *Pelvetia canaliculata* and *Ascophyllum nodosum* were also brought up, in addition to kelps like *Saccharina latissima* and *Laminaria digitata*. At the same time, uncertainty about classification, nomenclature and the differences between seaweed species, for example kelp and wrack, were prevalent. Despite the accessibility and the great abundance of this resource, the lack of common local terminology and the corresponding binomial nomenclature of the species used as feed make reporting a challenge, and indicate a rather scanty Norwegian coastal culture in this respect. Many farmers also reflected on the nutrients that the seaweeds might provide and how they might impact animal welfare and digestion.

The practice of using seaweeds for sheep fodder has declined dramatically over the past thirty years, after feed concentrate was introduced for Norwegian farming in the sixties. The structural rationalization towards larger and more efficient holdings therefore makes seaweed as a less favoured feed component. Our informants therefore see no feasible way of feeding large numbers of sheep indoor. In Røst, where winter temperatures are high, mature sheep are kept outdoors all winter even today. These animals are accustomed to eating seaweed on the beaches and find their own way to the shore during low tides, or at certain times of year. Outdoor grazing is however, challenged in some areas due to infrastructure (fences and roads), changing property regimes, and fragmentation of pastures which leads to less easy access to beaches. Nevertheless, the farmers clearly see the value of local protein sources like seaweeds as part of sheep's fodder intake. However, our informants have concerns about the sustainability and environmental impacts connected to large scale harvest of kelp forest.

> I am concerned over the kelp forest. Kelp forests are our rainforests and we cannot harvest it without being aware of the consequences. These forests are habitat for juveniles and small fish and their food. Today fishing of copepods for aquaculture threatens juveniles. Sea urchins have taken so much already. There is not enough for people too. (Fiskarbonde at Røst, Oct 2014).

The sheep farmers interviewed in Røst also have concerns about the practicalities and economics connected to large-scale use of this fodder resource. The agriculture industry and individual farmers are exposed to constant changes in economic, managerial, and climatic/environmental conditions. Although their sheep use the beach as pastures, the usage of these resources has not been modernized into present value chains and production lines. Today's farms are larger in scale and farmers perceive it as practically very challenging to feed large numbers of sheep with seaweeds. To include seaweed in future sheep fodder, large scale cultivation or harvest is therefore necessary. Concerns of the local fishermen and farmers over the ecological impacts, from cultivation, harvesting and/or collecting beach-cast seaweeds on coastal environment and fisheries require further studies.

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Management and previous sward composition influence the potential for establishing species-rich grassland

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Abstract

In the context of a land consolidation measure, plant-species rich grassland should be established on a former field. In an observational study it was investigated whether the site had potential for establishing species-rich grassland (comparison of sowing and natural establishment) and which impact two different management systems – exclusively grazing by cattle and mowing (first growth) combined with grazing – have on vegetation composition. After five experimental years, significant changes in plant composition caused by different establishing types were observed: The natural establishment resulted in higher proportions of grazing-tolerant plant species (related to accidental seeding of a seed-mixture for intensive use prior to the experiment). Except for differences in the abundance of some plant species, e.g. *Achillea millefolium* and *Lolium perenne* in the seeded treatments, having larger yield shares in the mown and grazed or in the grazed plots, respectively, the management had no significant influence on vegetation composition.

Keywords: grazing, mowing and grazing, vegetation composition

Introduction

Grazing animals are affecting biodiverse grassland by treading, forage selection, excretal deposition and their role in propagule dispersal (Rook and Tallowin, 2003). The heterogeneous sward structure, created hereby, is crucial for plant species richness (Schley and Leytem, 2004). Both, vegetation composition and sward structure were found to be important for e.g. insect diversity (Dumont *et al.*, 2007) and abundance (Jerrentrup, 2013). Grazing is required to maintain certain biotopes like *Mesobromion* with *Juniper*, but is often seen to be critical with respect to plant species diversity in typically mown habitats.

Materials and methods

The experimental site is located within a nature reserve in in the south of Baden-Wuerttemberg (county Ravensburg, Germany: 582 m above sea level). The mean annual temperature is 8.1 °C; the mean annual precipitation is 975 mm. The soil is a luvisol, the texture sandy loam and partly loamy sand. The site was used as arable land for growing grain. In 2008, a conventional grassland seed mixture was sown accidentally. In autumn 2009, the whole site was ploughed and harrowed. Half of the site was sown with a species-rich seed mixture (20 kg ha^{-1}: containing 54% forbs [*Achillea millefolium, Centaurea jacea, Cirsium oleraceum, Crepis biennis, Leucanthemum vulgare, Lychnis flos-cuculi, Plantago lanceolata, Polygonum bistorta, Prunella vulgaris, Ranunculus acris, Rumex acetosa, Salvia pratensis, Sanguisorba officinalis, Silene dioica, Tragopogon pratensis*], 30% grasses [*Alopecurus pratensis, Arrhenaterum elatius, Festuca pratensis, Holcus lanatus, Poa pratensis*] and 16% legumes [*Lotus corniculatus, Onobrychis viciifolia, Lotus uliginosus, Trifolium pratense*]) in the midst of October and rolled ('Cambridge-type roller'). The other half was left uncultivated. From 2010 onwards, both halves were grazed with Scottish Highland cattle or mown (first growth) and grazed (Table 1). The first use in the grazed plots was from the midst of May on, i.e. earlier than mowing (midst of June – beginning of July), to achieve a more homogeneous growth use.

Treatments were replicated only twice due to shortage of space. Plot size varied between 366 and 611 m^2, as the two replications differed in width. The whole experimental site had an area of 0.30 ha. Adjacent grassland was linked with the experimental plots, resulting in a size of 2 ha during the first growth and

Table 1. Treatments.

Treatments	Factor 1: plant establishment	Factor 2: management
1. 0-G	open soil (0)	grazing only (G)
2. 0-MG		mowing of first growth, then grazing (MG)
3. S-G	sowing of species-rich mixture (S)	G
4. S-MG		MG

2.2 ha (combined with mown and grazed plots) for following growths. The stocking density (4.0-7.1 livestock units ha^{-1}) and stocking period (10-49 days) varied between growths and years. For vegetation analyses, yield dry matter proportions of plant species were recorded on 24 m^2 (3 subplots of 8 m^2) per plot. Statistical tests were performed using data of 2014 and 2015 in univariate (R, Version 3.2.3; data were transformed, when the preconditions were not met) as well as in multivariate (redundancy and principle component analyses: Canoco for Windows Version 4.5, using default settings and log-transformed species abundances) repeated measurements analyses.

Results and discussion

No treatment effects on the yield proportion of grasses, forbs and legumes and total plant species number were observed (data not shown). Vegetation composition was affected by the method of grassland establishment (P=0.032) (Figure 1). Plant species, whose abundance was significantly correlated with the natural establishment were *Medicago lupulina*, *Trifolium pratense*, *Bellis perennis*, *Plantago lanceolata*, the agriculturally undesired gap closers *Cirsium arvense* and *Agropyron repens* (both <1% yield share in 2015) and species typical for intensively used grassland (*Dactylis glomerata*, *Taraxacum officinalis*, *Lolium perenne* and *Trifolium repens*). The latter presumably reestablished from the former seed mixture and led to a comparatively high tolerance of the sward against treading, grazing (Briemle *et al.,* 2002) and mowing (Briemle and Ellenberg, 1994; all parameters: P<0.001) as well as in negligible proportions of the undesirable plant species mentioned above. Plant species, whose abundance was significantly correlated to seeding were *Sanguisorba officinalis*, *Anthoxanthum odoratum*, *Rumex acetosa*, *Ranunculus acris* and the seeded species *Achillea millefolium*, *Alopecurus pratensis*, *Centaurea jacea*, *Cirsium oleraceum*, *Crepis biennis*, *Holcus lanatus*, *Leucanthemum vulgare*, *Lotus* spp., *Prunella vulgaris* and *Salvia pratensis* (t value biplot). The total yield share of sown species was 25-30% higher in seeded compared to uncultivated treatments (P<0.001).

The sown plant species *Lychnis flos-cuculi*, *Onobrychis viciifolia*, *Bistorta officinalis*, *Silene dioica*, *Tragopogon pratensis* did not establish in any of the treatments. This might have different reasons: high legume proportions (34-38%), found in all treatments, may have prevented their establishment (Bosshardt, 1998) or the sowing date might have been too late, resulting in winter killing of young seedlings. The sowing depth was between one and two cm. However, producer of species-rich seed mixtures recommend sowing onto the soil surface. The regions for which those seed mixtures are offered are relatively large, necessitating a wide range of site conditions presented by the plant species included. Considering this, it is obvious that on a plot with relatively homogeneous site conditions, not all of the sown plant species could find favourable conditions.

Overall, the number of sown species both in the sward of the sown and uncultivated treatment was nearly identical with the exception of *Cirsium oleraceum* that established only in the seeded plots. Some of the seeded species were presumably present in the prior seeding mixture, like *Alopecurus pratensis* and *Festuca pratensis*. Other species were most likely spread by the animals that grazed all plots in the second and later growth. The dispersal distances of plant species are often rated as low (Bosshardt, 1998) – with the

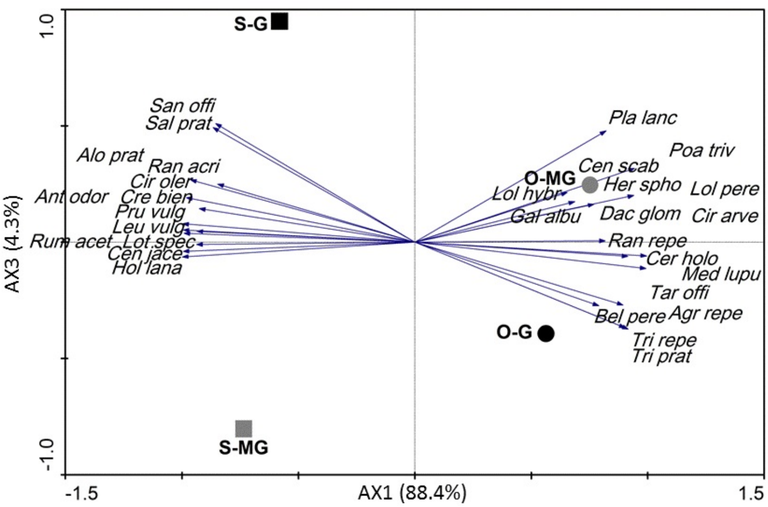

Figure 1. Principal component analysis of 2015. Shown are plant species (as arrows) and treatments (for description see Table 1). Plant species: Agr repe = *Agropyron repens*; Alo prat: *Alopecurus pratensis*; Ant odor = *Anthoxanthum odoratum*; Bel pere = *Bellis perennis*; Cen jace = *Centaurea jacea*; Cen scab = *Centaurea scabiosa*; Cer holo = *Cerastium holosteoides*; Cir arve = *Cirsium arvense*; Cir ole = *Cirsium oleraceum*; Cre bien = *Crepis biennis*; Dac glom = *Dactylis glomerata*; Gal albu = *Galium album*; Her spho = *Heracleum sphondylium*; Hol lana = *Holcus lanatus*; Leu vulg = *Leucanthemum vulgare*; Lol hybr = *Lolium hybridum*; Lol pere = *Lolium perenne*; Lot spec = *Lotus spec.*; Med lupu = *Medicago lupulina*; Pla lanc = *Plantago lanceolata*; Poa triv = *Poa trivialis*; Pru vulg = *Prunella vulgaris*; Ran acri = *Ranunculus acris*; Ran repe = *Ranunculus repens*; Rum acet = *Rumex acetosa*; Sal prat = *Salvia pratensis*; San offi = *Sanguisorba officinalis*; Tar offi = *Taraxacum officinale*; Tri repe = *Trifolium repens*; Tri prat = *Trifolium pratense.*

exception of wind-dispersed seeds, therefore Poschlod *et al.* (1998) emphasize the role of grazing animals for grassland seed dispersal. Management did not affect vegetation composition. Minor differences were at best noticeable in the seeded treatment. *Achillea millefolium* was more abundant in the mown and grazed treatment (P<0.001; 7 vs 4%), while *L. perenne* achieved larger yield proportions in the grazed treatment (9 vs5%; P=0.002).

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Effect of management practice on floristic composition of lowland permanent grasslands

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Abstract

In Serbia permanent grasslands represent a significant source of animal feed. However, in most regions the animal production and grazing, and thus the productivity and investment in grasslands, are on a very low level. This research aims to analyse impact of nitrogen fertilization and harrowing on lowland permanent grassland (Vojvodina province, Serbia) in order to improve floristic composition and thus yield and quality. Research was carried out during 2012-2015 in Vojvodina province, Serbia, on permanent grassland which was mainly used for sheep grazing. Trial included two nitrogen rates, namely 40 kg ha^{-1} and 80 kg ha^{-1}, divided on plots with and without harrowing, as well as control treatments. Floristic composition was mostly composed of species from families *Poaceace* and *Asteraceae* with a small number and presence of legume species. The number of species changed in the third and fourth years of the trial. *Cynodon dactylon* L., *Lolium perenne* L., *Festuca* sp., were significantly higher on plots with the higher nitrogen rate, while on the control the most frequent spp. were *Trifolium campestre* Schreb. and *Medicago lupulina* L. Harrowing, on the other hand, had smaller impact on improvement of permanent grassland.

Keywords: cutting regime, fertilization, floristic composition, harrowing, permanent grassland

Introduction

Permanent grasslands are defined as areas used to grow grasses or other herbaceous forage naturally (self-seeded) or through cultivation (sown) and that have not been included in the crop rotation of the holding for five years or longer (EU Commission Regulation No 1307/2013). In Serbia agricultural land covers 5.1 million ha, or 58%, of which 0.83 million ha are permanent grasslands (SORS, 2013). Lowland permanent grasslands are dominated in the north part of the country, in Vojvodina province on different types of soil, which are mostly not suitable for crop production (Eric *et al.*, 2007). Their productivity and quality has decreased over the years as a consequence of decreased animal production and grazing, the absence of any application measures and unfavourable weather conditions. Different researches have shown that species diversity declines when grasslands are abandoned or as a result of land use during their history (Lindborg and Eriksson, 2004). Fertilization, moderate grazing or mowing, harrowing and pesticide application are all management practices which can stop grassland degradation and improve its diversity and yield. The aim of this research was to analyse impact of nitrogen fertilization and harrowing on lowland permanent grassland in order to improve its floristic composition and thus its yield and quality.

Materials and methods

Research was carried out during 2012-2015 in Vojvodina province, Serbia, on one permanent grassland which was mainly used as a pasture. The soil was calcareous gleyed chernozem, with pH (H_2O) 8.06, pH (KCl) 7.37, N 0.24%, P_2O_5 50.16 mg 100 g^{-1} and K_2O 38.87 mg 100 g^{-1}. The area was divided into treatments organized in a randomized block design with four replications. Plot size was 12 m^2. Treatments included two nitrogen rates, namely 40 kg ha^{-1} and 80 kg ha^{-1}, divided on plots with and without harrowing, as well as control treatments. Harrowing was performed by tractor aggregated with harrow. Fertilizer application was done manually each year at the beginning of growing period. The area was surrounded by a fence and used for cutting. In all years, samples were taken before the first cut and plants were identified and sorted by hand into legumes, grasses, weeds and others herbaceous plants. Precipitation and temperature data for all years are given in Table 1.

Table 1. Mean monthly temperature and precipitation data for hydrological years 2011-2015 (RHMZ, 2012-2016).

	Mean temperature (°C)					Mean precipitation (mm)				
	2011/2012	2012/2013	2013/2014	2014/2015	LTA[1]	2011/2012	2012/2013	2013/2014	2014/2015	LTA
Oct	10.7	13.6	13.6	14.1	11.6	35.0	49.0	66.2	65.0	48.0
Nov	2.8	10.0	8.4	9.2	6.2	2.0	36.0	41.0	6.0	50.0
Dec	4.3	1.0	1.6	4.6	1.4	49.0	55.0	1.3	65.0	49.0
Jan	1.7	3.1	4.3	3.1	-0.3	45.0	60.5	24.0	59.0	39.0
Feb	-4.7	5.8	6.7	3.1	1.6	66.0	47.2	10.0	66.0	34.0
Mar	8.1	6.0	10.2	7.7	6.3	4.0	73.0	50.0	58.0	38.0
Apr	13.0	13.4	13.5	11.7	11.7	83.0	35.8	49.0	15.0	47.0
May	17.5	18.1	16.1	17.8	17.0	53.0	118.1	203.0	193.0	61.0
Jun	23.0	20.4	20.4	20.5	20.0	27.0	125.7	38.0	27.0	88.0
Jul	25.3	22.2	22.3	24.6	21.7	48.0	34.1	143.0	2.0	67.0
Aug	24.6	23.2	21.5	24.3	21.2	4.0	26.7	80.0	99.0	57.0
Sep	19.8	16.0	18.3	20.2	16.9	13.0	107.8	88.0	53.0	48.0

[1] LTA = long term average.

Results and discussion

The floristic composition of analysed grassland was very diverse in terms of species and plant number and it was mostly composed of species from family *Poaceace* and *Asteraceae* (Table 2). Due to very extreme weather conditions in 2012 the numbers of grasses and particularly legumes were reduced, and at the beginning of 2013 we identified only one legume species. Based on graphic interpretation of the preliminary results, the number of species changed in the third and fourth years of the trial when the number of legumes (mostly *Trifolium campestre* Schreb. and *Medicago lupulina* L.) was higher on control treatments regardless of the harrowing. The number of grass species, in particular *Cynodon dactylon* L., *Lolium perenne* L., *Festuca* sp., was significantly higher on plots with the higher nitrogen rate. Fertilization had an impact on floristic composition by reducing the number of species but emphasizing productivity level through higher number of grasses (Figure 1).

Table 2. Identified species divided on legumes, grasses, weeds and other herbaceous plants (2012-2015).

Plants group	Identified species
Legumes	*Medicago lupulina, Trifolium campestre, Tryfolium hybridum, Trifolium repens, Vicia* sp.
Grasses	*Festuca rubra, Cynodon dactylon, Bromus mollis, Bromus sterillis, Lolium perenne, Agropyron repens, Hordeum murinum, Dactylis glomerata, Festuca ovinna, Digitaria sanguinalis, Poa pratensis, Festuca arundinacea, Setaria viridis, Sorghum halepense*
Weeds and other herbaceous plants	*Achillea millefolium, Bellis perennis, Cichorium intybus, Sonchus arvensis, Taraxacum officinalis, Carduus acanthoides, Erigeron canadensis, Matricaria chamomile, Ambrosia artemisifolia, Capsella bursa-pastoris, Sisymbrium officinale, Stellaria media, Euphorbia helioscopia, Erodium cicutarium, Geranium dissectum, Lamium amplexicaule, Papaver rhoeas, Plantago lanceolata, Plantago major, Convolvulus* sp., *Veronica hederifolia*

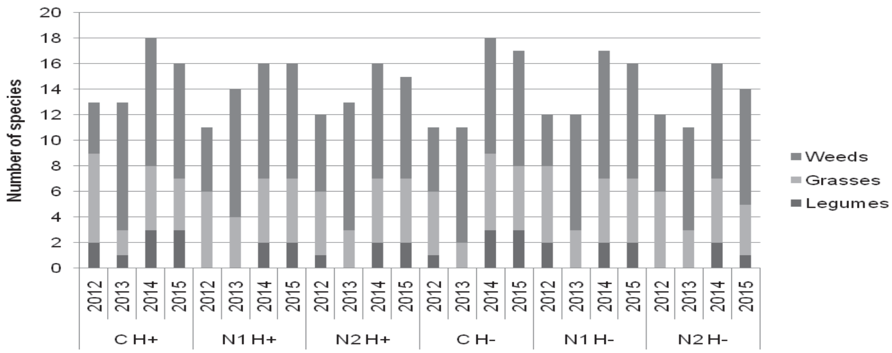

Figure 1. The number of species from three groups-legumes, grasses, weeds and other herbaceous plants at the beginning of growing period in each year during 2012-2015. C H+ = control with harrowing, N1 H+ = 40 kg ha^{-1} with harrowing, N2 H+ = 80 kg ha^{-1} with harrowing, C H- = control without harrowing, N1 H- = 40 kg ha^{-1} without harrowing, N2 H- = 80 kg ha^{-1} without harrowing.

Conclusions

It is implied that nitrogen fertilizers change the floristic composition of lowland grassland by favouring grasses and other herbaceous plants, but also by reducing the number of legumes. Harrowing, on the other hand, may serve as measurement for improving soil structure, species diversity and hay quality, but it requires more time and larger plot size to accomplish grassland improvement.

Acknowledgements

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Carbon partitioning to roots of maize hybrids differing in maturity group

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Abstract

Silage maize production is assumed to negatively affect soil C content. This study quantified the impact of hybrid/sampling date on the above- and belowground dry matter and C accumulation, carbon input and root turnover during the growing season. Hybrid/harvest date had no impact on gross root biomass and C input, which amounted to 1.2 Mg C ha^{-1}. Root turnover was estimated at 50% until harvest. In contrast to the roots, the stubble biomass increased when harvest was delayed, resulting in higher total C input for later harvests.

Keywords: *Zea mays* L., hybrid, yield, root-to-shoot ratio, soil organic carbon

Introduction

Soil organic carbon (SOC) is an important determinant of soil quality and ecosystem functioning, with roots and stubble contributing most to SOC accumulation (Kätterer *et al.*, 2011). According to the German carbon balancing approach, silage maize (*Zea mays* L.) production is associated with a depletion of SOC (VDLUFA, 2014), which can be compensated by organic fertilisation and catch crops. Successful catch crop establishment in northern Germany, requires timely harvest of the preceding maize (Van Dam, 2006). Early harvest, however, may limit root and stubble biomass accumulation during the post-silking phase (Mackay and Barber, 1986). The objective of the present study was to analyse the dynamics of root biomass accumulation of two maize hybrids differing in maturity group, which were harvested successively to estimate the C input to the soil as well as the root turnover during the growing season.

Materials and methods

A 2-year field experiment was conducted at the research farm 'Ostenfeld' in Schleswig-Holstein, northern Germany. The site is characterised by a silty-sandy soil and mild-humid climate with an average annual precipitation sum of 847 mm and a mean annual temperature of 8.9 °C. The experimental setup was a three-factorial (year, hybrid/harvest date, N fertilisation) randomised block design with three replications and a plot size of 51 to 76.5 m^2. Treatments comprised two years (2012, 2013), two N fertiliser levels (N0 and N180), four consecutive harvest dates, i.e. 10 Sept, 20 Sept, 30 Sept, 15 Oct (hd1-d4), with two hybrids nested in harvest dates: early-maturing 'Suleyka' (S210/K240) and mid-early 'Ronaldinio' (S240/K240). Suleyka was harvested at hd1 and hd2, and Ronaldinio at hd3 and hd4. Maize was sown in the last ten days of April after cultivation (mould-board plough) and seedbed preparation (rotary harrow) with a plant density of 10 plants m^{-2} in rows 0.75 m apart. All plots received basal dressings of phosphorus (75% as banded starter), potassium and magnesium applied at 124 (P_2O_5), 250 (K_2O) and 37 (MgO) kg ha^{-1}, respectively. If fertilised with N (calcium-ammonium-nitrate), broadcast shortly after sowing, the target value of 180 kg N ha^{-1} was adjusted by taking soil mineral N into consideration. In addition to the final harvests by plot harvester, aboveground dry biomass (AGB) accumulation was quantified by 5 manual harvests, i.e. at silking and two samplings each before and after until harvest dates. The stubble dry matter (DM) was sampled manually only at hd1 and hd3 by cutting 10 stubbles per plot to soil surface. Carbon and N content of shoot samples was determined by near infrared spectrometry (NIRS). Belowground gross biomass accumulation (BGB) was quantified by the ingrowth-core technique (Steingrobe *et al.*, 2001), where soil-filled mesh bags were inserted into the soil to a depth of 30 cm for 4-week periods

(ST: short-term), then removed and roots are washed out. Additionally, longer periods of 8 (MT: mid-term), 13 (LT: long-term) and 16 weeks (W: whole), were included in the second experimental year and compared to the cumulated ST-intervals, to allow root turnover estimation. Root N and C contents were determined by the DUMAS combustion method (Vario Max CN, Elementar Hanau, Germany). Analyses of variance were calculated using 'R' software by assuming year, hybrid/harvest date and N fertilisation as fixed and block as random. Repeated measurements effects were accounted for by assuming appropriate correlation structures. Multiple comparisons of means were conducted by linear contrasts ($P \leq 0.05$).

Results and discussion

Both the above- and belowground DM (AGB, BGB) sampled during the growing season until harvest were affected significantly by the interaction of year × hybrid × N × sampling date ($P \leq 0.01$). As expected, AGB increased significantly until harvest. The hybrid showed only an effect in fertilised treatments; with a 25% lower AGB of Suleyka at hd1 than Ronaldinio in 2012, while in 2013 a 22% higher AGB was found. BGB increased significantly until harvest, but the growth rates ceased after silking (103 DAS), which is in accordance to results reported by Niu *et al.* (2010), finding a net reduction of root biomass in the post-silking phase. The effects of hybrid and N fertilisation were minor. Pooled over N, hybrids and years, the BGB at harvest was 2.5 Mg DM and 1.2 Mg C ha^{-1} and the N accumulation in roots averaged 54.4 kg N ha^{-1} with a C/N ratio of 24. The root-to-shoot ratio (R/S) was affected significantly by the interactions of year × sampling date ($P \leq 0.05$) and hybrid × sampling date ($P \leq 0.01$) and decreased during the growing seasons from 1.3 to 0.17 or 1.2 to 0.2 for Ronaldinio and Suleyka, respectively averaged over years (Figure 1A). Consistently, R/S dropped until 103 DAS (silking) in both years and again until harvest, except for Suleyka in 2012. At harvest, R/S deviated significantly between 2012 and 2013 (0.24 vs 0.13) averaged over hybrids. Neither hybrids nor N fertilisation levels differed significantly at the different sampling dates, but averaged over years and N fertilisation, R/S of Suleyka (0.22) exceeded that of Ronaldinio (0.17) due to increased yield with delayed harvest. The duration of the ingrowth-core

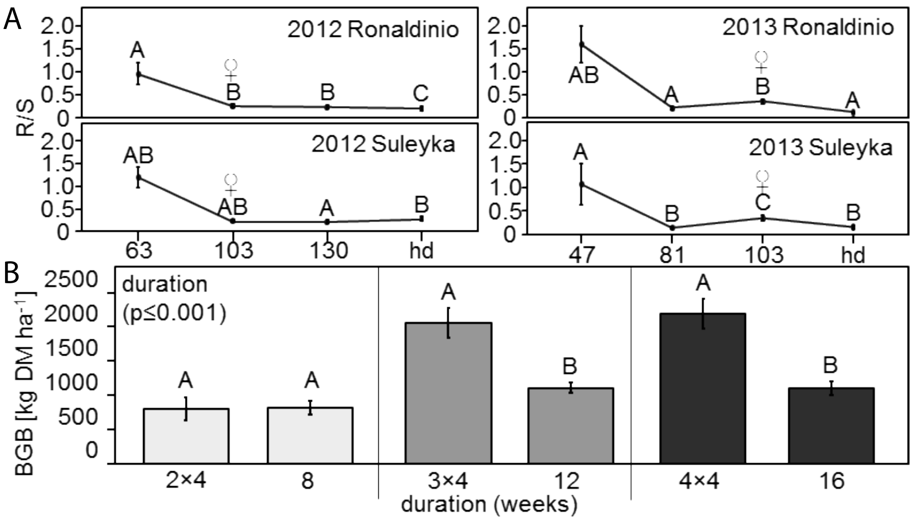

Figure 1. (A) Changes in root-to-shoot ratio (R/S) during the growing season (DAS: days after sowing), pooled over N treatments and (B) impact of ingrowth core period on BGB determined until the juvenile, silking stage and harvest dates. ST = short-term (4 weeks), MT = mid-term (8 weeks), LT = long-term (13 weeks) and W = whole (16 weeks): the figure following the abbreviation provides the number of intervals, e.g. ST3 = 3×4 weeks. Capital letters denote differences between sampling dates and between ingrowth periods in (A) and (B) ($P \leq 0.05$); ♀ indicates the date of silking. Bars represent standard error of the mean.

periods had a significant impact on BGB ($P \leq 0.001$), as revealed by ANOVA. ST2 and MT showed no significant differences, which indicates that root gross DM determination with higher temporal resolution did not have a negative impact (Figure 1B). In contrast, longer ingrowth-core periods LT and W resulted in a 54 and 50% lower BGB compared to ST3 and ST4, respectively, which corresponds to results reported by Chen *et al.* (2015). Aboveground DM yield at final machine harvest (stubble not included) was affected by harvest date ($P \leq 0.001$), showing a significant yield loss of 11 to 13% at hd1 compared to delayed harvest (hd2-hd4), which achieved an average yield of 18.7 Mg DM ha^{-1}, and did not differ significantly. Stubble DM was influenced significantly by the interaction of year × harvest date × N ($P \leq 0.01$), resulting in a lower stubble DM for hd1 compared to hd3 (average: 0.98 vs 1.3 Mg DM ha^{-1}). As stated by Menichetti *et al.* (2015), a fraction of 30 and 15% of below- and aboveground plant C inputs resides in SOC. Accordingly, the C input residing in soil in the present study varied between 0.51 (hd1) and 0.55 Mg C ha^{-1} (hd2-hd4).

Conclusions

The C input by roots (480 g C kg^{-1} DM) was on average 1.2 Mg C ha^{-1}, without any impact of hybrid/harvest date, assuming discontinued root accumulation after hd1. Delaying harvest increased stubble C from 0.46 to 0.61 Mg C ha^{-1}, and resulted in a total C input of 1.66 (hd1) and 1.81 Mg C ha^{-1} (hd3). The results further reveal that root C input may be substantially underestimated when sampling roots by augers, which does not allow taking root turnover into consideration. From a yield perspective, there seems no need to further delay harvest beyond the second decade of September, nor will this have adverse effects on SOC either.

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Short-term effects of cutting frequency and organic fertilizer on species composition in semi-natural meadows

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Abstract

Permanent hay meadows in montane regions serve as advertising subjects for food industry, considered as highly diverse, flowering and healthy forage in ruminant production. On the other hand, farmers tend to intensify their cutting and fertilising regime on accessible sites, which may result in low diverse, uniform stands of high fodder quality for ruminants. Traditionally, Austrian mountain farmers mow their meadows two times a year without periodic re-sowing. We tried to analyse changes in vegetation under different management regimes in a triannual case study consisting of a block design with 30 sub-plots on two comparable mesophilic meadows of the *Arrhenatheretalia* R. Tx. 1931 in the Eastern Alps. The effect of management intensity (2, 3, 4 cuts; manure vs slurry) was tested for species coverage, e.g. dominants like *Trisetum flavescens, Dactylis glomerata, Trifolium repens, Taraxacum officinale* and *Achillea millefolium*. The results show changes in abundance of widespread meadow species after three years due to management intensification. Stolon- and rosette-forming plants profited from increased management whereas tall tuft grasses lost coverage. While the less foraging grass *Poa trivialis* accelerated, on the other hand valuable species as *Poa angustifolia* declined in abundance. Hence, there will be limits to intensification on permanent grasslands, dependent on site condition and species composition.

Keywords: *Arrhenatheretalia*, cutting frequency, organic fertilizer, meadow species

Introduction

In Central Europe, semi-natural meadows in mountain regions that are unsuitable for crop production, offer an important source for grazing and conservation of silage and hay in livestock breeding. Austrian grasslands contribute to about half of agricultural area (1.38 m ha^{-1}). Therefrom, 25 m ha^{-1} can be described as traditionally used semi-natural grassland with a maximum of two cuts or grazing terms per year. However, the greater part, 0.48 m ha^{-1}, belongs to improved meadows, used more than 3 times per year with adapted fertilization (BMLFUW 2015). During the last centuries, a clear management intensification occurred due to socio-economic changes. Meadows having appropriate climatic and soil conditions in lowland regions and in mountain areas were transformed to uniform swards consisting only of a few productive species (Bassler *et al.,* 2011; Dietl, 1995; Isselstein *et al.*, 2005). Inclination limited management intensity during past centuries, this might change with availability of modern machinery along with growing farm size due to structural changes in agriculture. In this case study, the floristic changes of representative *Arrhenatheretalia* stands in Styria, Austria, due to management intensification were analysed.

Materials and methods

The experiment was carried out in the Upper Styrian Pöls valley on the crystalline bedrock of the `Niedere Tauern', Austria. After a vegetation survey in 2008, the two most homogenous meadow fields were selected at 920 and 980 m a.s.l., both facing 230° SW and characterised by an average inclination of 25%. Soil type was a typical cambisol structurally dominated by sandy loam. In 2009 the following average soil parameters were observed: pH ($CaCl_2$) 5.7±0.17 mg kg^{-1} (site 1) and 5.9±0.15 mg kg^{-1} (site 2); P (CAL) 36±8 mg kg^{-1} (site 1; low P availability) and 71±17 mg kg^{-1} (site 2, high P availability);

K (CAL) 133±41 mg kg^{-1} (site 1, good K availability) 271±68 mg kg^{-1} (site 2, high K availability). Annual mean temperature at the farm ranges from 6.5 to 8 °C and annual precipitation from 850 to 1040 mm from 2009 to 2011. The original mesophilous grassland referred to the association *Cardaminopsido halleri-Trisetetum flavescentis* (Bohner and Sobotik, 2000), typical for mountainous hay meadows on crystalline bedrock. No sowing of cultivars was performed within 15 years before the experiment. 30 plots á 4 m^{-2} were arranged at each site in a block design. The six treatment factors included 2, 3 and 4 cuts, in combination with application of either slurry or manure. Before each mowing we recorded cover percentage of each species. Statistical analysis were implemented with proc MIXED of SAS 9.2 (P<0.05) for species specific mean coverage on 1 to 3 of June in 2012. The resulted model LS-means were displayed as percent coverage together with standard error of the model (SEM). Test of pairwise differences were arranged with Tukey-Kramer and significant differences are shown with different lower-case letters. For a detailed experiment description see Starz *et al.* (2015).

Results and discussion

How the treatments influenced mean coverage of dominant species, is shown in Table 1. The tuft grass *Festuca pratensis* lost abundance in intense mowing regime, in contrast to *Lolium perenne*. Perennial ryegrass, a grass of short stature was more susceptible to intensive management. Meadow fescue was the single grass which seems sensitive to slurry application, as already mentioned in Dietl *et al.* (1998). Management had no impact on coverage of *Dactylis glomerata*, which can be explained by its life span, which was estimated to be six to eight years (Schmitt, 1995). There was no effect of the treatments on coverage of *T. flavescens* in this study, but former findings showed a decline of this species due to increased cutting regime (Angeringer *et al.,* 2011). The herbs *Crepis biennis, Taraxacum officinale* and *Achillea millefolium* showed declining cover values with increasing management intensity, especially the subterranean shoots forming *A. millefolium*. *Poa angustifolia*, a valuable grass with subterranean runners, significantly lost cover with intense mowing. In 2012, aggregate taxon *P. pratensis* was divided into small-leafed *P. angustifolia* and broad-leafed *P. pratensis*. This became necessary, because regional ecotypes of the latter were not susceptible to intensification as mentioned before (Angeringer *et al.,* 2011). On the other hand, cultivars of *P. pratensis* are used in seed mixtures for intensively managed meadows and pastures in Austria (Starz *et al.*, 2010). *Poa trivialis* is a grass which was able to form numerous aboveground down-

Table 1. Influence of treatments on cover percentage (%) of species with high continuity (>90%) in 1-3 June 2012 (n=60).

Parameter	Cutting frequency					Organic fertilizer			
	2	3	4			Slurry	Manure		
	%	%	%	SEM	P-value	%	%	SEM	P-value
Lolium perenne	11b	14b	19a	1.3	0.0008	15	15	1.1	0.8410
Poa trivialis	7a	10a	22b	1.5	<0.0001	14	12	1.3	0.1279
Poa angustifolia	22a	21a	16b	1.2	0.0010	19	21	0.9	0.0479
Poa pratensis	1a	1a	2a	0.4	0.1442	2	1	0.3	0.5621
Trisetum flavescens	16a	12a	14a	1.3	0.0899	15	14	1.2	0.3485
Dactylis glomerata	4a	3a	4a	0.4	0.1303	4	4	0.4	0.1067
Festuca pratensis	3ab	4b	2a	0.4	0.0165	2	3	0.3	0.0345
Crepis biennis	10a	7b	5b	0.6	<0.0001	7	7	0.5	0.4261
Taraxacum officinale	7a	6b	6b	0.4	0.0096	6	7	0.3	0.0605
Achillea millefolium	15a	16a	8b	0.9	<0.0001	13	13	0.7	0.9473
Trifolium repens	12a	13a	15a	1	0.0938	12	14	0.9	0.0673

[1] Tested with GLM Proc. Mixed, post-hoc test Tukey-Kramer in different lower-case letters.

bending shoots after the first mowing. It increased in treatments with three and four cuts a year through capturing free gaps by those organs. There were no changes in coverage of the single legume in this study, *Trifolium repens* though there was a slight preference for intense cutting and manuring.

Conclusions

After three years of different management regime, we recorded significant changes in coverage of the most frequent meadow species due to cutting regime, but few responses to the type of organic fertiliser. Species in permanent grassland stands competed for nutrients and gaps. Most of the well-adapted hay meadow species decline in coverage due to intensification, e.g. *P. angustifolia*. Of the valuable forage crops, only *L. perenne* increased in coverage after intensification. Thus, the occurring gaps are susceptible for settlement of less palatable species as *P. trivialis*. This field experiment demonstrated the development of montane *Arrhenatheretalia* meadows from species-rich stands to fragile swards consisting of undesirable species if mowed more often without reseeding.

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Flower-rich habitat formation for wild pollinators in intensive agriculture lands

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Abstract

The experiment was carried out in 2014 at the Institute of Agriculture (Dotnuva) and at the Joniskelis Experimental Station (Joniskelis) of the Lithuanian Research Centre for Agriculture and Forestry with the aim of determining annual and perennial species combinations suitable for pollinators and evaluating the effects of field margins on intensively used lands. Floral sward margins were established on the edges of large intensive farming lands (>5 ha). The margins were seeded with four plant combinations: perennial legumes, annual dicotyledons, natural grassland and perennial grasses. The most intensive flowering period was in June and July in all swards combinations. *Onobrychis viciifolia* Scop., *Trifolium rubens* L., *Sinapis alba* L., *Galium boreale* L. were the earliest flowering species, while *Medicago sativa* L., *Fagopyrum esculentum* Moench. and *Phacelia tanacetifolia* L. had a longer flowering period. It was found that the floral swards margins had a positive impact on the yields of legume crops grown near margins.

Keywords: legume, grass, pollinators, bloom

Introduction

Most European land is devoted to agriculture, therefore modern agricultural intensification, increased fertiliser and pesticide use will inevitably contribute to the loss of semi-natural habitats. The significant negative effect on the densities of different kinds of pollinators, including wild bees, due to habitat loss and fragmentation of natural material has been documented by Brittain *et al.* (2010). Intensive land reclamation of farming areas clear-cuts of forest edges and large agricultural spaces in Lithuania have led to the deterioration of the bio-ecological state of the landscape (Jonaitis *et al.*, 2002). Researchers have already shown that new-alternative food, as well as creation of micro-habitat fragmentation can increase the abundance and diversity of pollinators (Carré *et al.*, 2009). The goal of this research was to set flowering annual and perennial herbaceous species combinations in field margins, which might be suitable to attract pollinator insects in intensive farming conditions of Central Lithuania.

Materials and methods

The experiments of floral swards margins were carried out in 2014 in two places: at the Institute of Agriculture (Dotnuva) and at the Joniskelis Experimental Station (Joniskelis) of the Lithuanian Research Centre for Agriculture and Forestry, with the aim of determining annual and perennial species combinations suitable for pollinators and evaluating the effects of field margins on intensively used lands. Experimental sites were different in landscape homogeneity. In Joniskelis, the diversity of insect species was noticeably smaller and less natural habitat fragments were available for pollinators compared to Dotnuva. Floral sward margins were established on the edges of large intensive farming lands (>5 ha). The margins were seeded with four plant combinations: perennial legumes (*Trifolium pratense* L., *Onobrychis viciifolia* Scop., *Medicago sativa* L., *Trifolium repens* L., *Lotus pedunculatus* Cav.) 20 kg ha^{-1}, annual dicotyledons (*Helianthus annuus* L., *Fagopyrum esculentum* Moench, *Borago officinalis* L., *Sinapis alba* L., *Phacelia tanacetifolia* L., *Linum usitatissimum* L., *Lupinus luteus* L., *Melilotus albus* L., *Trifolium resupinatum* L.) 33 kg ha^{-1}, natural grassland (grasses: *Poa palustris* L., *Poa compressa* L., *Corynephorus canescens* L. *etc.;* perennial legumes: *Trifolium rubens* L., *Medicago lupulina* L., *Vicia cracca* L. *etc*.; other species: *Centaurea* *jacea* L., *Agrimonia eupatoria* L. *etc.*) 16 kg ha^{-1} and perennial grasses as control treatment (*Festuca arundinacea* Schreb., *Festuca pratensis* Huds., *Festuca rubra* L., *Phleum pratense* L., *Dactylis glomerata* L., *Agrostis capillaris* L.) 20 kg ha^{-1}. The seeds for natural grassland mixtures were collected in semi-natural meadows. We assessed the pollinators' presence visually throughout the flowering period, from early June to late August, in five 0.25×0.25 cm plots and counted four times the number of open flowers in the same plots. Pollinators (*Hymenoptera* species: various bees, wasps and hornets) surveys were conducted four times throughout the blooming period (Saville *et al.*, 1997). During the investigation period, the weather conditions were favourable for flying insects: it was dry (with the exception of July) and warm (especially in the second half of the summer). The research data was statistically processed by a one-factor analysis of variance using the program package Selekcija (Tarakanovas and Raudonius, 2003).

Results and discussion

Micro-habitat margins differed by plants blooming duration and abundance. The overall flowering period continued from early June to August. In Dotnuva, the annual dicotyledons mixture was the earliest in flowering (Table 1). In the Northern part of Lithuania (Joniskelis), the phenology was delayed about 1-2 weeks compared to Central part (Dotnuva): the highest amount of flowers was assessed in early July, in Dotnuva, and in late July, in Joniskelis. The most intensive blooming was registered in the perennial legumes mixture, in Dotnuva, and in the annual dicotyledons one, in Joniskelis. The natural grassland mixture showed a low amount of blooms. The earliest species were *O. viciifolia*, *T. rubens*, *S. alba*, *P. media*, *G. mollugo* and *G. boreale*. In the second part of the summer, *H. annuus* and *Me. albus* started to flower when most of the other species finished their blooming. *M. sativa, F. esculentum* and *P. tanacetifolia* displayed the longest flowering period. Joniskelis site is characterised by low forest coverage and the creation of a huge agricultural space, which implies bad biocenological conditions. Pollinators were less abundant in the perennial grasses margin, compared to the other mixtures (Figure 1). Pollinator visits was highest from early June until middle July (Dotnuva) and from late June until late July (Joniskelis).

Table 1. Numbers of flowers in the different field margins (units per m^2).[1]

Seed mixture	Dotnuva (Open flowers, units per m^{2})				Joniskelis (Open flowers, units per m^{2})			
	Early June	Early July	Late July	Late August	Early June	Early July	Late July	Late August
Perennial grasses	0	415.2	420.8	0	10.0	8.0	38.4	0
Perennial legumes	13.6	596.8**	499.2**	92.0*	0.4**	85.6**	412.8**	80.0**
Annual dicotyledons	132.8**	272.0	396.8**	7.2	7.2	236.0**	836.8**	50.4**
Natural grassland	0	176.0	119.2**	32.0*	6.6	91.2**	95.2	23.2

[1] * and ** are significant at P<0.05 and P<0.01 levels of probability, respectively, using Duncan's multiple range test.

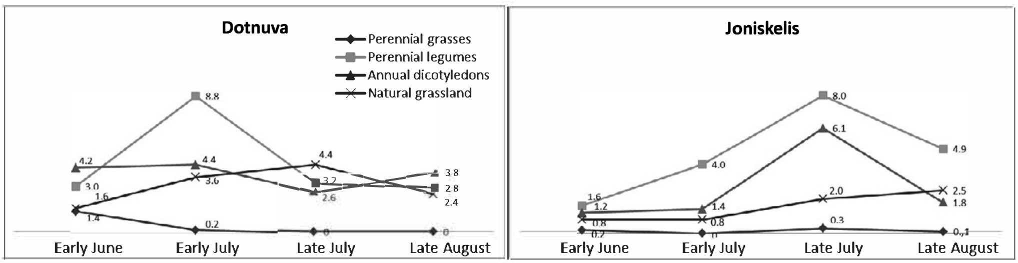

Figure 1. Abundance of *Hymenoptera* pollinator species in the different field margins (units per m^2).

Perennial legume sward resulted in being most attractive for pollinators. It was also found that the field margins had positive impact trends on the yields of legume crops grown nearby (data not presented).

Conclusions

In field margins, the flowering time depended on climate (which differs between the two sites). In each site, it depended on the species composition of the mixtures (with early or late flowering species). Different types of bees, bumble bees, hornets and wasps mostly attended the margins where blooming was most intensive, namely in perennial legumes and annual dicotyledons.

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Root mass of differentially defoliated patches on a long-term grazing experiment

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Abstract

Grazing affects biodiversity and habitat structure, due to defoliation, trampling pressure and the shifting of nutrients. In low-intensity grazing systems, selective grazing leads to the formation of areas that are more frequently defoliated (short patches) than others (tall patches), which is called patch grazing. The different patch types are supposed to have different aboveground biomass productivities. The stocking rate determines the ratio of short and tall patches on a pasture. In a long-term low-intensity cattle grazing experiment with three different stocking rates (target sward heights: 6 cm (moderate), 12 cm (lenient) and 18 cm (very lenient)), the root and shoot biomass of two different patch types (short and tall) was analysed. Due to frequent defoliation we assumed that the root mass is lower in short patches than in tall patches. In contrast to the assumption, the root mass of tall patches was significantly lower than the root mass in short patches. The adverse effect of frequent defoliation on root mass might have been superimposed by a nutrient effect as the nitrogen availability in tall patches appeared to be higher than in short patches.

Keywords: defoliation, root biomass, patches, stocking rate

Introduction

Grazing affects plant growth especially due to the removal of photosynthetic tissue (Ferraro *et al.*, 2002). In low-intensity grazing systems, patch grazing (Dumont *et al.*, 2007) leads to the formation of areas that are more frequently defoliated (short patches) than others (tall patches). In a long-term low-intensity cattle grazing experiment with three different stocking rates (target sward heights: 6 cm (moderate), 12 cm (lenient) and 18 cm (very lenient)), the root and shoot biomass of two different patch types (short and tall) was analysed. We assumed that the frequent loss of aboveground biomass within the short patches would also affect the belowground biomass. Due to a more even nutrient distribution under more intensive grazing (Moir *et al.*, 2010), it is possible that short patches within the moderate stocking rate have higher nutrient concentrations than the short patches of the lenient stocking rates. A lack of nutrient forces the plant to increase the root mass (Poorter *et al.*, 2000), this leads to the assumption that root mass of the short patches differs between the different stocking rates. We hypothesized (1) that short patches have a lower root mass than tall patches and (2) that root mass of short patches under moderate stocking rate is lower than that of short patches under lenient and very lenient stocking rate.

Materials and methods

The experiment was conducted on a long-term low-intensity continuous cattle grazing experiment at the experimental farm in Relliehausen (51°46'N, 9°42'E, 250 m a.s.l.), of the University of Göttingen. The average annual temperature is 8.2 °C and the average annual rainfall is 879 mm. There has been neither fertilizer nor herbicide application for at least 20 years. Since 2005, three stocking rates defined by paddock target sward heights have been compared: moderate, lenient and very lenient stocking, with 6, 12 and 18 cm compressed sward height (CSH), respectively, measured via be-weekly rising-plate-meter measurements (Castle, 1976). Target sward heights were kept constant in a put-and-take system. Since 2013 two different patch types have been defined according to their CSH, with short: CSH ≤33rd percentile and tall: CSH ≥67th percentile of 450 bi-weekly CSH measurements. The experiment was composed of three randomized blocks, comprising a total of 9 paddocks with 1 ha each. In 2013 and 2014 aboveground biomass of two plots per patch type and paddock was harvested at two times per year (May and October) in a stratified sampling design, comprising 72 plots in total. Per plot eight soil cores with a diameter of 1.64 cm each were taken to a depth of 20 cm. Roots were washed, dried and weighed to determine root dry mass. The software R 3.2.2 (R Core Team, 2015) was used for statistical analyses. Due to the nested experimental design, data were fit with linear mixed effects models. 'Year', 'date', 'stocking rate', 'patch type' and their interactions were set as fixed factors and 'paddock' nested in 'block' as random factors. Data were averaged over the two plots per patch type and paddock. Minimum adequate models were determined via Akaike's Information Criterion (AIC).

Results and discussion

The present results show that root/shoot ratio is significantly higher in short than in tall patches (Table 1), reflecting that short patches are defoliated frequently and tall patches are defoliated rarely. Due to the frequent loss of photosynthetic tissue, root dry matter of short patches was expected be lower than that of tall patches. Instead, we found a higher root mass in short compared to tall patches (Table 1). Furthermore, a lower root biomass in short patches under moderate stocking in comparison to the more lenient stocking rates could not be found. Thus, both hypotheses have to be rejected. However, root dry mass was lower in 2013 than in 2014, which can presumably be explained by less rainfall in the first year. Our results are in contrast to studies of Ferraro *et al.* (2002) and Thornton *et al.* (1996), who found a reduction in root mass due to defoliation. It is likely that the adverse effect of frequent defoliation on root mass might have been superimposed by a nutrient effect. A depletion of soil nutrients within short patches and a dislocation of these nutrients to tall patches during the long-term cattle experiment may be

Table 1. Root mass (g dry matter m^{-2}) and root/shoot ratio of different patch types in May and October 2013 and 2014. Means and standard errors of three stocking rates and three replications as well as *F* and *P*-values of effects included in the final linear mixed effects models are shown.

Year	Date	Patch type	Root mass	Root/shoot ratio
2013	May	Short	34.0±3.9	0.75±0.19
		Tall	30.6±5.3	0.09±0.02
	Oct	Short	28.8±4.1	0.73±0.19
		Tall	21.1±2.8	0.08±0.01
2014	May	Short	18.3±2.5	0.17±0.03
		Tall	19.6±2.1	0.04±0.00
	Oct	Short	16.9±3.0	0.29±0.05
		Tall	13.9±1.5	0.05±0.01
Results of linear mixed effects models				
Year			F=12.3, P<0.001	F=54.8, P<0.001
Date			*F*=1.6, *P*=0.2105	*F*=3.9, *P*=0.0544
Patch			F=4.9, P=0.0308	F=105.3, P<0.001
Grazing			*F*=0.7, *P*=0.5600	Not included in final model
Year×patch			*F*=1.8, *P*=0.1872	*F*=8.0, P=0.0063
Date×patch			Not included in final model	*F*=1.9, *P*=0.1765
Year×date			*F*=1.7, *P*=0.2022	Not included in final model
Year×grazing			*F*=1.4, *P*=0.2525	Not included in final model
Date×grazing			*F*=2.4, *P*=0.1012	Not included in final model
Year×date×grazing			*F*=2.1, *P*=0.1393	Not included in final model

assumed, as Ebeling *et al.* (2016) found short patches to be less productive than tall patches at the same experimental site. Plants are able to adapt their carbon allocation to changing above- or belowground resources. A lack of nutrients, as it may exist in short patches of this experiment, would force the plant to increase belowground biomass in order to capture those plant growth limiting resources (Poorter *et al.*, 2000).

Conclusions

The study indicates a significantly reduced root mass in tall patches and no variation in root mass due to different stocking rates. The reason for this effect could be a depletion of soil-nutrients within the short patches and a dislocation of these nutrients to the tall patches. A closer look at the nitrogen fluxes and related productivity is expected to improve the understanding of root biomass formation in grazed heterogeneous pastures.

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Impact of the sheep grazing season on xerothermic grassland sward utilisation

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Abstract

The study objective was to assess the impact of the sheep grazing season on xerothermic grassland sward utilisation in the context of biodiversity. Phytosociological studies of the plant cover in xerothermic grassland of the *Festuco-Brometea* class were conducted in 2015, in three Natura 2000 areas (Stawska Góra PLH060018, Zachodniowołyńska Dolina Bugu PLH060035 and Kąty PLH060010) in eastern Poland. The grazing of 40 native sheep of the Świniarka and Poland Lowland Sheep breeds was conducted with two rotations in the first two areas, and, in the third area, with one rotation but two plant communities: xerothermic grassland and fallow land as potentially restorable grassland. Grazing was conducted on an area of 12.10 ha. After the grazing, the degree of xerothermic grassland sward utilisation was analysed and the seasons of grazing were assessed in the context of conserving environmentally valuable habitats. The utilisation of xerothermic grassland sward in the context of the first regrowth was significantly the largest in the case of grazing in May and the lowest in October. Sward utilisation was decreasing significantly with the postponing of the grazing commencement date.

Keywords: *Festuco-Brometea* class, Natura 2000 site

Introduction

Calcareous xerothermic grasslands of the *Festuco-Brometea* class are among Natura 2000 sites facing the greatest threat of extinction in Europe. They are a sanctuary for rare and endangered flora and faunal species, but abandoning the use of these grasslands leads to undesirable habitat changes (Bornkamm, 2006). Semi-natural xerothermic grasslands are formed as a result of long-lasting agricultural land use. Generally, these communities are rarely used today for agricultural purposes. The biggest threats to xerothermic grasslands include the lack of grazing, secondary succession, invasive alien species and native expansive species as well as accumulation of dead organic matter, which leads to succession changes in the natural habitat (Kulik *et al.*, 2015). In view of the decreasing numbers of farm animals, including sheep, the protection of xerothermic grasslands is a difficult challenge requiring precise planning of grazing, particularly in the case of restorable initial swards. The study objective was to assess the impact of the sheep grazing season on xerothermic grassland sward utilisation in the context of its protection.

Materials and methods

Phytosociological studies of the plant cover in xerothermic grasslands of the *Festuco-Brometea* class were conducted in 2015, in three Natura 2000 sites (Stawska Góra PLH 060018 – Staw, Zachodniowołyńska Dolina Bugu PLH 060035 – Gródek and Kąty PLH 060010 – Kąty II) in eastern Poland. Stawska Góra, located in Staw village, is both a flora reserve (4.9 ha) and a Natura 2000 site and constitutes a unique sanctuary for many rare, relict plant Natura 2000 species, including *Carlina onopordifolia* (code 2249), endemic to Ukraine and Poland (Melnyk, 2013). Xerothermic grasslands in Gródek represent a small part of the PLH 060035 Nature 2000 site (1,556.11 ha) while in Kąty II it represents 23.98 ha of the PLH 060010 site. Phytosociological relevés were taken using the Braun-Blanquet method. The grazing of 40 native sheep of the Świniarka and Poland Lowland Sheep breeds was conducted with two rotations in the first two areas while, in the third area, with one rotation but two plant communities: xerothermic grassland

and fallow land as potentially restorable grassland. Grazing was conducted in an area of 12.10 ha (10.22 ha on xerothermic grassland). After the grazing, the degree of xerothermic grassland sward utilisation was analysed on 4 m^2 plots (quadrat method) estimating abundance of vegetation ingested and non-ingested by sheep with an accuracy of 5%. The obtained degrees in 5 repetitions were subjected to the ANOVA analysis complemented by the Tukey test (P<0.05). The vascular plant nomenclature according to Mirek *et al.* (2002) and the classification of communities according to Matuszkiewicz (2008) were used.

Results and discussion

Phytosociological studies revealed the presence of valuable natural habitats. Habitat 6210-3 flowering xerothermic grasslands from the *Cirsio-Brachypodion pinnati* alliance (*Festuco-Brometea* class) predominates in three Natura 2000 areas. In Gródek, the occurrence of *Thalictro-Salvietum pratensis* and *Origano-Brachypodietum pinnati* associations was found. Furthermore, in Staw and Kąty, there are patches of thickets from the *Rhamno-Prunetea* class (Table 1) that pose a grave threat to xerothermic grasslands (Bornkamm, 2006), particularly in Stawska Góra where *C. onopordifolia* grows, a very rare and endemic species (Melnyk, 2013; Kulik *et al.*, 2015). In Kąty, it is a priority habitat with significant *Orchidaceae* sites with the predominant association *Inuletum ensifoliae* and undeveloped, initial swards with the predominance of *Anthyllis vulneraria* and characteristic species of the *Festuco-Brometea* class. Fallow land is covered by a community with *Elymus repens* corresponding to the *Convolvulo arvensis-Agropyretum repentis* association with characteristic species from ruderal communities, xerothermic grasslands and thermophilous ecotones.

The utilisation of xerothermic grassland sward was significantly the largest in the case of grazing in May (98%) and the lowest in October (3%). It should be noted that in Gródek in the August/September period, and in Staw in the September/October period, part of the area was grazed in the first and another part in the second rotation. This influenced the preferences of the sheep and the degree of sward utilisation (Table 2). A similar situation occurred in Kąty, where the utilisation of fallow land vegetation (48 and 26%) was significantly lower than the utilisation of xerothermic grassland sward (92 and 62%, respectively); both areas were grazed simultaneously. During the July/August grazing, the sheep preferred the sward of xerothermic grassland over fallow land vegetation despite the close proximity of the latter.

Conclusions

The utilisation of xerothermic grassland sward in the context of the first regrowth was significantly the largest in the case of grazing in May and the lowest in October. Sward utilisation was decreasing significantly with the postponing of the grazing commencement date. In Kąty, the utilisation of

Table 1. Phytosociological classification of xerothermic grassland

Nature 2000 site/area	Vegetation type	Dominant species
Stawska Góra PLH 060018/Staw	All. *Cirsio-Brachypodion pinnati*	*Carlina onopordifolia, Brachypodium pinnatum, Melampyrum arvense*
	Cl. *Rhamno-Prunetea*	*Rhamnus catharticus, Cornus sanguinea, Prunus spinose, Viburnum opulus*
Zachodniowołyńska Dolina Bugu	Ass. *Thalictro-Salvietum pratensis*	*Elymus hispidus subsp. hispidus, Salvia pratensis*
PLH 060035/Gródek	Ass. *Origano-Brachypodietum pinnati*	*Brachypodium pinnatum*
Kąty PLH 060010/Kąty II	Ass. *Inuletum ensifoliae*	*Inula ensifolia, Linum flavum*
	initial sward	*Anthyllis vulneraria*
	community with *Elymus repens*	*Elymus repens, Hieracium piloselloides, Taraxacum officinale, Achillea millefolium*

Table 2. Degree of xerothermic grassland sward utilisation in 2015.[1]

Vegetation type	Nature 2000 site/area	Season/rotation	Grazing area (ha)	Sward utilisation degree (%)
xerothermic grassland	Stawska Góra PLH 060018/Staw	4-30 May/1st	1.39	98[a]
		30 September – 27 October / 2nd	1.39	62[c]
		30 September – 27 October / 1st	0.24	3[e]
	Zachodniowołyńska Dolina Bugu PLH 060035/Gródek	1-30 June / 1st	1.00	94[ab]
		28 August – 29 September / 2nd	1.00	82[b]
		28 August – 29 September / 1st	0.60	9[e]
	Kąty PLH 060010/Kąty II	1 July – 3 August / 1st	0.72	92[ab]
		4-27 August / 1st	1.18	62[c]
fallow land		1 July – 3 August / 1st	0.82	48[c]
		4-27 August / 1st	1.88	26[d]

[1] Different letters indicate significant differences $P<0.05$.

xerothermic grassland sward was significantly higher than the utilisation of fallow land vegetation. In the context of preserving biodiversity, including zoochory, i.e. the influence of sheep on the seed dispersal of xerothermic species, it should be concluded that July should be the latest time of grazing commencement due to the progressive lignification of vegetation.

Acknowledgements

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Foraging activity of red deer population on restored mid-forest meadows

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Abstract

Availability of meadow habitats in the territory of free-living red deer (*Cervus elaphus*) may reduce the negative impact of these animals on forests and arable crops. The aim of this study was to evaluate the foraging activity of red deer on restored mid-forest meadows. In 2014-2015 two experiments located in North-West Poland were carried out, that investigated the effects of different restoration methods (full tillage, including sowing of three types of seed mixtures: M1 – 'Blühende Wildäsung', M2 – 'Weidgreen Hochwildweide', M3 – the author's seed mixture, and overdrilling) vs control surface without restoration on the foraging activity of red deer, based on sward browsing intensity, sward yield and quality. Most of the grazing activity and the highest yield and herbage quality were found within areas restored by the full tillage method combined with sowing of specialised seed mixtures. The results indicate that the areas restored by full tillage and sowing of specialised seed mixtures stimulated grazing by red deer and increased herbage yield and quality.

Keywords: mid-forest meadows, red deer, restoration, yield, herbage quality, sward browsing intensity

Introduction

Red deer (*Cervus elaphus*) are considered a keystone species for the functioning of agro-forestry systems and have a great impact on the structure of the ecosystem. The increasing size of the red deer population causes more and more damage to agricultural and forest crops (Marchiori *et al.*, 2012). A limitation of this problem has become an important task for game species management. One possible solution could be to relocate the red deer foraging activity from cultivated areas. Mid-forest meadows (grassland areas surrounded by forest which are grazed by wild animals and/or mown 1-2 times per year for conservation of hay or silage) are habitats that can become attractive feeding grounds for red deer. However, because of their forest proximity and ecotone character, mid-forest meadows are often of low economic and environmental value (Sankey, 2012). Red deer, as all ruminant herbivores, choose feeding patches based on the quantity and quality of available forage (Gerhardt *et al.*, 1993). Therefore, increasing productivity of mid-forest meadows by combining various sward treatments and sowing of properly composed seed mixtures may possibly control their food base. The aim of this study was to evaluate red deer foraging activity on restored mid-forest meadows, based on sward browsing intensity in relation to herbage yield and quality.

Materials and methods

The data collection on the foraging activity of a free-living red deer population on restored mid-forest meadows was carried out in 2014 and 2015. For this purpose, two mid-forest meadows in the Polanów Forest District (54°10' N, 16°78' E) were used as experimental sites. The control areas without restoration (C) on each meadow were compared to the differently restored areas: (1) full tillage combined with sowing of three types of seed mixtures: M1 – 'Blühende Wildäsung'; M2 – 'Weidgreen Hochwildweide' (both commercially available); and the author's seed mixture – M3 (composed of 23 species: 7 grasses, 5 legumes and 11 herbs); (2) overdrilling using perennial ryegrass and white clover (O).

Sward browsing intensity in all experimental areas (0.3 ha each) was assessed by ranking the percentage of grazed area (0 – no foraging evidence; 1 – 0-25%; 2 – 25-50%; 3 – 50-75%; 4 – 75-100% of grazed surface) in 9 separate plots (2×2 m squares). Sward yield was estimated by placing three caged grazing exclosures made of 2×2 m wire mesh within each experimental area. Dry matter (DM) yield was measured based on manually harvested sward samples collected inside the cages. Results are given as an average yield of four harvesting terms, according to the standard management schedule of mid-forest meadows in the Polanów Forest District (sward harvest for hay production in June and October before the end of the vegetation season). The collected herbage was dried in an oven at 60 °C for 48 hours. Chemical composition of the herbage in each of the experimental areas was determined according to the Weende method. Crude ash (CA), crude protein (CP), ether extracts (EE) and crude fibre (CF) were analysed by means of near infrared reflectance spectroscopy; and nitrogen free extractives (NFE) were calculated as g per kg of dry matter. The significance of differences between restoration methods was estimated by the Kruskal-Wallis test (one-way ANOVA on ranks) using Agricolae library for R (version 3.2.3).

Results and discussion

The highest average DM yield per cut was observed in the area restored using the full tillage method and seed mixture M1 (3,774 kg ha^{-1}) or M2 (2,496 kg ha^{-1}) (Figure 1). Lower DM yield was observed on M3 sown areas (2,011 kg ha^{-1}). The yields per cut within the overdrilled and control areas were not statistically different (1,692 kg ha^{-1} and 1,647 kg ha^{-1}, respectively).

The sward browsing intensity analysis indicated that the areas restored using full tillage became so attractive to the red deer that they focused about 90% of their foraging activity on these areas (Figure 2). The most preferred was mixture M1 – 46.5% of this treatment area was grazed. The areas sown with mixtures M2 and M3 were less attractive – 31.5 and 31.3% of grazed surface, respectively. Overdrilling did not have a statistically significant effect – only 4.1% of the area was browsed, compared to 3.4% in the control areas.

The restoration of mid-forest meadows exerted a significant impact on herbage quality, particularly on CP, EE and CF. The best nutritional quality was recorded in the samples taken from the areas restored by the full tillage method (Table 1): CP content ranged from 171 to 191 g kg^{-1} DM depending on the seed mixture M1-M3, and was significantly higher than in the samples from the overdrilled and control areas (151.6 and 150.4 g kg^{-1}, respectively). The effect of the restoration of mid-forest meadows on the content of CA and NFE in sward was not observed.

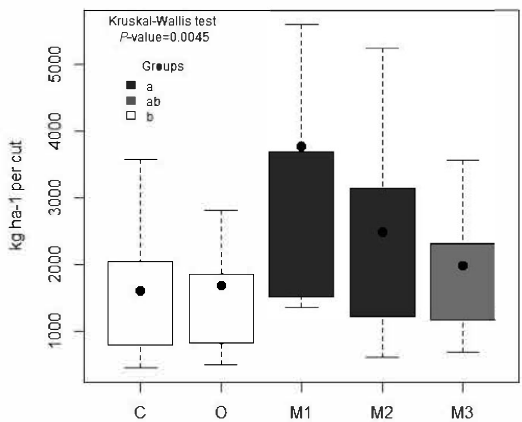

Figure 1. Effect of mid-forest meadow restoration on dry matter yield.

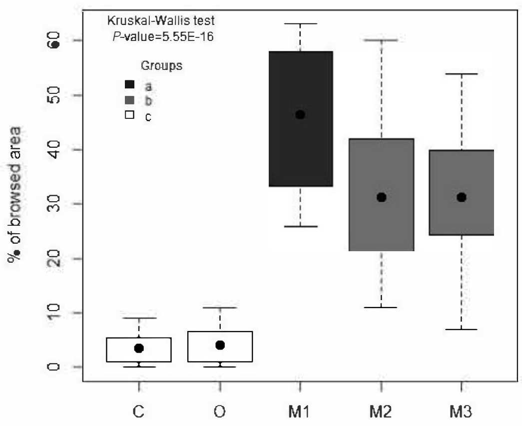

Figure 2. Effect of mid-forest meadow restoration on sward browsing intensity.

Table 1. Effect of mid-forest meadow restoration on the chemical composition of herbage (means 2014-2015 in g kg^{-1} of dry matter).[1]

Restoration method	Crude ash	Crude protein	Ether extracts	Crude fibre	Nitrogen free extractives
Full tillage + M1	58.0a	190.7a	32.7a	232.0ab	486.6a
Full tillage + M2	65.6a	172.3a	32.8a	223.9b	505.4a
Full tillage + M3	60.8a	170.5a	31.1ab	221.6b	516.0a
Overdrilling	54.8a	151.6b	28.6bc	255.6a	509.4a
Without restoration	54.1a	150.4b	28.1c	257.9a	509.5a
P-value	0.7239	0.0041	0.0108	0.0118	0.8948

[1] Values marked with the same letter are not significantly different.

The results of our study confirm the statement by Langvatn and Hanley (1993) that, while feeding, red deer select 'most of the best and least of the worst but some of everything'. The foraging activity of red deer was observed in all experimental treatments, but it was significantly lower in the areas with low sward yield and low quality of mid-forest meadows. Langvatn and Hanley (1993) suggest that protein content in the herbage is a better indicator of forage attractiveness for red deer than productivity. In our study, the highest quality of herbage (as indicated by a high CP) was observed within areas restored using the full tillage method, and correlated with the highest sward yield.

Conclusions

The restoration of mid-forest meadows using full tillage combined with sowing of specialised seed mixtures positively affected both herbage yield and quality, which leads to greater foraging activity of free-living red deer. The sward browsing intensity of animals was significantly lower within the overdrilled and the control areas.

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A common shoot developmental framework for perennial legume species with contrasting morphogenesis

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Abstract

Little is known about the elementary traits which make temperate forage legumes differ so substantially in their growth habit and morphologies. In the present study, we compared the patterns of shoot organogenesis and of organ growth of six contrasting forage species (namely alfalfa, birdsfoot trefoil, sainfoin, Kura clover, red clover and white clover) during their vegetative phase. An experiment was carried out over two years in a greenhouse under non-limiting water and soil nutrients. Phytomer initiation appeared driven by temperature and highly deterministic in all the species in the absence of competition for light. The temporal sequence of organ growth differed between species. However, organ growth was highly coordinated within a phytomer in all the species, and was independent of the position and axis order when expressed in phyllochronic time. By contrast, organ dimensions at maturity were dependent on phytomer position, and followed a regular function of the rank for all the organs. Overall, a very similar developmental pattern was followed by all the species, but they differed greatly in the absolute values taken by their developmental traits.

Keywords: legumes, alfalfa, clover, morphogenesis, development

Introduction

Many perennial forage legume species are grown in temperate grasslands with a common general purpose: to provide high-quality N-rich food for ruminants and reduce the need for nitrogen fertilizers. In spite of their close phylogenetic relationship, they display a very large range of architecture, growth habits and ecological niches (Forde, 1989; Thomas, 2003). Identifying the major traits differentiating the performance of legume species along environmental gradients (e.g. soil water availability), or with respect to different companion species, is essential to target relevant traits in breeding programs and to improve the assembly of multi-species grassland. Yet, little is known in this group of species concerning the major traits promoting leaf area expansion, height increment, and ultimately dry matter production. Existing developmental frameworks usually focus on a particular species (e.g. white clover – *Trifolium repens*, Gautier *et al.,* 2000; *Medicago truncatula*, Moreau *et al.,* 2006; alfalfa – *Medicago sativa*, Baldissera *et al.,* 2014; red clover – *Trifolium pratense*, Van Minnebruggen *et al.,* 2011). To our knowledge, no attempt to develop a unified quantitative description of the morphogenesis of these species has been made. In this study, we selected six contrasted temperate legume species amongst the most widely grown and challenged the hypothesis of a common framework of vegetative development under non-limiting growing conditions.

Materials and methods

Two independent experiments were carried out in a heated greenhouse at INRA's Lusignan station, France, from February to April in 2014 and 2015. Plants of alfalfa (cv. Timbale), white clover (cv. Giga), red clover (cv. Formica), sainfoin (*Onobrychis viciifolia* cv. Canto), birdsfoot trefoil (*Lotus corniculatus* cv. Leo) and Kura clover (*Trifolium ambiguum* cv. Sevanskij) were grown until the end of the vegetative stage in 10 l pots in a randomized blocks design. Seedlings were transplanted in the pots after germination in Petri dishes (48 h at 25 °C in the dark). Plants were ferti-irrigated daily with complete nutrient solution (Gastal and Saugier, 1986, 300 ml day^{-1}) and were grown at low density (2 plants m^{-2}). Supplemental light was provided 16 h day^{-1} using 400 W HQI lamps. Plant measurements included a counting of the visible leaves on the primary shoot and branches, and organ (i.e. leaf, petiole, stipule, internodes) length on a sample of phytomers. All statistical analyses were performed with the R software version 3.1.2 (2014).

Results and discussion

Phytomer productions on the primary axis and its branches were linear functions of thermal time (Figure 1). Primary leaf appearance rate ranged from 2.1 10^{-2} leaves growing degrees days $(GDD)^{-1}$ (phyllochron = 76.8 GDD) in white clover to 3.2 10^{-2} leaves GDD^{-1} (phyllochron = 30.8 GDD) in alfalfa. No significant differences between the rate of development of branches at different positions on the primary axis were found in any of the six species; however, the rate of development of branches differed from the rate of the primary axis in alfalfa, kura clover and sainfoin (ANCOVA, $P<0.05$). Similar linear relationships between leaf appearance and thermal time have been established across several species including alfalfa (Louarn *et al.,* 2007; Baldissera *et al.,* 2014). However such regularity between ranks was unexpected (Moreau *et al,*. 2006; Louarn *et al.*, 2007). This finding makes it easy to predict phytomer production by individual shoots using simply two phyllochrons (one for the primary axis and one for the branches).

The expansion of organs (leaflet lamina, petiole, and internode) was characterized by two values: its timing of appearance (t_{50}, defined as the time at which the organ reached 50% of its final length; in phyllochronic time relative to leaf appearance) and the timespan needed for the organ to complete 95% of its expansion (d_{95}, in phyllochronic time). Both values for every species are summarized in Table 1. Within a species, these parameters were not significantly different between phytomers at different topological positions. However, they differed strongly between species (ANOVA, $P<0.01$). There was some consistency in species ranking (e.g. alfalfa and birdsfoot trefoil both always 5th or 6th; white clover always 1st, 2nd or 3rd), but otherwise no consistent correlation between the two parameters or between organs.

Organ dimensions at maturity always appeared as regular functions of the rank of their phytomer (data not shown). Despite differences between the maximum values reached in the two experiments, relative profiles were consistent (ANCOVA with year as a categorical independent variable and rank as a covariate,

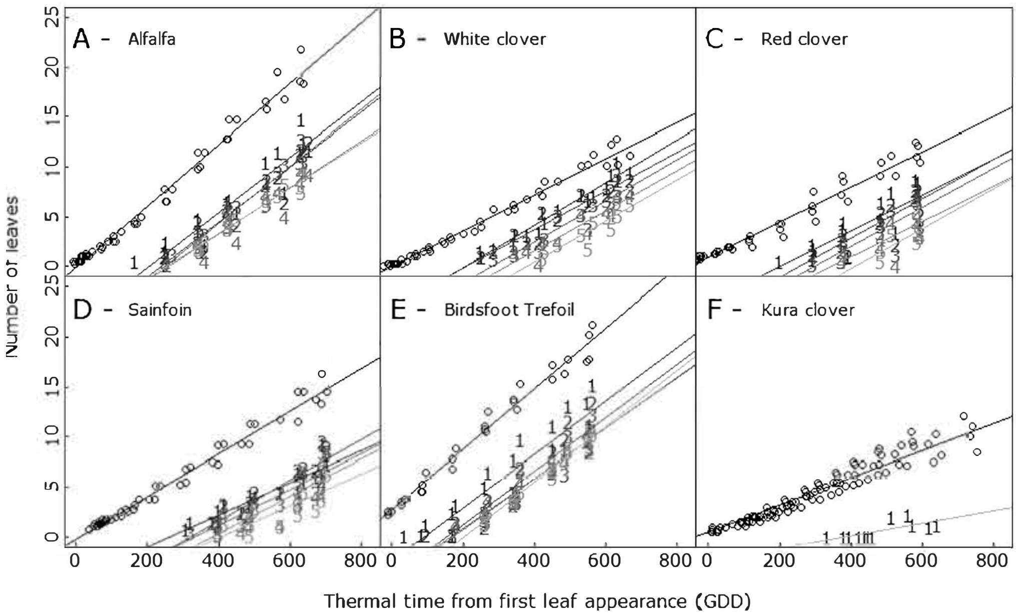

Figure 1. Leaf appearance rate on the seminal axis (open circles) and first 5 ramifications (digits from 1 to 5 in increasingly light shades of grey) of six forage legume species in the vegetative stage. The solid lines are linear regressions fitted to the data.

Table 1. Expansion parameters of the organs of six legume species (mean ± standard deviation).[1,2]

Species	Leaf lamina		Petiole		Internode	
	t_{50}	d_{95}	t_{50}	d_{95}	t_{50}	d_{95}
White clover	0	2.5±0.6^{a}	0.7±0.1^{ab}	2.6±0.4^{a}	0.0±0.1^{a}	1.4±0.3^{a}
Kura clover	0	2.7±0.5^{a}	0.9±0.2^{c}	2.5±0.4^{a}	1.7±0.4^{c}	3.2±1.2^{b}
Red clover	0	4.3±0.4^{b}	0.8±0.3^{bc}	4.5±1.2^{b}	1.8±0.5^{c}	3.4±0.3^{bc}
Sainfoin	0	4.4±0.4^{b}	0.5±0.2^{a}	2.5±0.5^{a}	1.0±0.1^{b}	2.8±0.4^{b}
Alfalfa	0	5.4±0.6^{c}	1.3±0.2^{e}	4.6±0.5^{b}	2.2±0.4^{c}	4.4±0.8^{c}
Birdsfoot trefoil	0	7.2±1.3^{d}	1.1±0.3^{d}	4.9±1.0^{b}	1.8±0.3^{c}	4.0±0.4^{c}

[1] d_{95} = duration of 95% of the expansion around the inflexion point of a logistic curve fitted to the data; t_{50} = delay between phytomer appearance and the inflexion point of the expansion curve, all values in phyllochrons.

[2] Superscript letters show homogeneous groups of species for each parameter (Newmann-Keuls tests after ANOVA).

performed separately on two linear sections of the profiles, P>0.05). This suggests that there is a strong species-dependent component to the patterns of organ dimensions along the stems (Allsopp, 1967).

Conclusions

All aspects of plant phytomer demography as functions of thermal time (up to the first order of ramification), and organ size as functions of phyllochronic time and position, appeared deterministic under non-limiting growing conditions. Unified parameters describing the components of leaf area expansion (i.e. leaf number and leaf size) and plant height were identified for the vegetative phase of contrasting forage legumes. Species differed greatly in the absolute values taken by their developmental traits, resulting in contrasting morphologies.

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The importance of spring and autumn grazing for seedling establishment in semi-natural grasslands

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Abstract

The diversity of available regeneration niches is generally considered a key factor for plant species diversity in semi-natural grasslands. The traditional management of hay meadows in Scandinavia included grazing in autumn and occasionally also in spring. In this study, I used a seeding experiment to examine the effects of spring and autumn grazing on seedling establishment in *Conopodium majus*, *Pimpinella saxifraga*, and *Knautia arvensis* in a hay meadow with traditional management. The experiment included four different combinations of autumn and spring grazing and three levels of disturbance in the bottom layer in a factorial design. The effects of grazing were highly species-specific and seasonally dependent. Autumn grazing had a clear positive effect on seedling establishment in *Pimpinella*. The results suggest that both removal of regrowth in the field layer and an increased frequency of gaps in the bottom layer (dominated by bryophytes) contributed to the positive effect of autumn grazing. In fact, no *Pimpinella* seedlings were present in plots without grazing or disturbance. In *Conopodium*, spring grazing alone or combined with autumn grazing resulted in the highest plant establishment at the end of the experiment one year after germination. In contrast to the other species, *Knautia* responded weakly to grazing. The results demonstrate that grazing in spring and autumn can have positive effects on seedling establishment in hay meadows and should therefore be considered as part of the management strategy in species-rich hay meadows.

Keywords: semi-natural grassland, grazing, traditional management, seedling establishment

Introduction

Species diversity in semi-natural grasslands is often limited by seedling recruitment, which in turn is determined by seed availability and safe sites for germination and establishment (Jongejans *et al.,* 2006). Large herbivores can favour seedling recruitment by reducing aboveground competition and litter accumulation, and by increasing the frequency of bare soil through trampling (Watt and Gibson, 1988). These changes improve light at the soil surface and thereby enhance successful germination and seedling establishment in many species (Bullock *et al,*. 1994, Lennartsson and Oostermeijer, 2001, Vandvik and Goldberg, 2006).

The main objective of this study was to estimate the impact of sheep grazing in either spring or autumn, or both, on seedling establishment in hay meadows. Seeds of three perennial grassland forbs were sown in an experiment which included four grazing treatments: (1) a control without grazing; (2) grazing in autumn; (3) spring; and (4) a combination of both autumn and spring. To examine the contribution of different disturbance regimes to the overall effect of grazing, the effect of biomass removal in the field layer and disturbance in the bottom layer were estimated within each grazing treatment.

Materials and methods

The study site was Havrå (60°29' N, 5°31' E) on the island Osterøy on the west coast of Norway, which is part of a protected cultural heritage site. The climate is oceanic, with an average annual precipitation is 2,225 mm, and every month having >100 mm. The experiment was conducted in an old meadow with a 25° slope, a south eastern aspect and intermediate soil moisture. The meadow is part of a larger area with semi-natural grassland and is still subject to the traditional management. The use of this meadow illustrates how traditional management can influence plant population dynamics. The traditional use includes sheep grazing in spring and autumn and mowing and hay making at the end of June or early July, though in some years the meadow was also mown a second time in August.

Three forbs, all characteristic of semi-natural grassland were studied: *Conopodium majus*, *Knautia arvensis* and *Pimpinella saxifraga*. To test the effects of grazing and disturbance regimes on seedling establishment, I performed a sowing experiment with four grazing regimes and three levels of disturbance. The experiment included eight blocks and 12 plots of 1×1 m within each block. In each plot there were three subplots of 0.2×0.2 m per species in which the three levels of disturbance were randomly applied (i.e. a split-plot design). The three levels of disturbance were: (a) removal of field layer, (b) removal of bottom layer, and (c) a control without disturbance (except from grazing). The grazing regimes included a control without grazing (1), grazing in autumn (2), spring (3) or both autumn and spring (4). Sheep grazed the meadow in spring (March 20 – May 20), and in autumn (September 20 – November 10). The stocking rate was 5-12 ewes ha^{-1} and the number of sheep was adjusted during the grazing periods to have a sward height of about 4-8 cm. Exclosures (1×1 m) of wire-netting regulated the grazing of individual plots.

Seeds of a single species were sown in each sub-plot, avoiding the outer 2 cm, and in autumn of the same year the seeds were harvested. I used 200 seeds per sub-plot for *C. majus* and *P. saxifraga*, and 100 seeds for *K. arvensis*. Seeds from local populations were sown in autumn 2003 and seedlings were counted in all sub-plots three times during a period of 1.5 years. Seedling establishment in each species was analysed separately using the number of seedlings per 100 seeds sown as the independent response variable. Because the response variables were proportions, generalized linear models with a logit link function and binomial distribution were used. Grazing, disturbance and census were considered as fixed effects and block as a random effect in the analysis.

Results and discussion

The effects of different grazing regimes were species-specific (Figure 1), whereas disturbance had consistently positive effects on seedling numbers in the three species. In *P. saxifraga,* autumn grazing had a positive effect on seedling establishment compared with the control and with spring grazing whereas spring grazing tended to have a negative effect on seedling establishment. The combination of spring- and autumn grazing did not affect seedling establishment when compared to the control without grazing. In *C. majus* spring grazing reduced seedling establishment. Plots with spring grazing and the combination of autumn and spring grazing had substantially less seedlings *C. majus* at the first census compared to autumn grazing and the control without grazing. There was no effect of grazing in autumn compared to control plots without grazing. The effect of spring grazing on *C. majus* is complex as grazing in spring actually improved survival of established juvenile individuals.

K. arvensis displayed low levels of seedling establishment and only 1.8% of the sown *Knautia* seeds resulted in a seedling the first spring. Grazing regime had a significant effect on seedling establishment in the analysis of variance (P=0.026). Plots with autumn grazing and the combination of autumn and spring grazing tended to have more seedlings than control plots without grazing but the differences were not significant at the end of the experiment.

For all species included in the study, disturbance stimulated seedling appearance, and the combination of disturbance in both the field and bryophyte layer gave significantly more seedlings compared to disturbance in the field layer alone. This pattern was present across all grazing regimes. Still, the magnitude of the effect of disturbance in the field and bottom layer depended on the grazing regime. The observed interactions between grazing and experimental disturbances indicate that autumn grazing is of particular importance for creating gaps for seedling establishment in the sward.

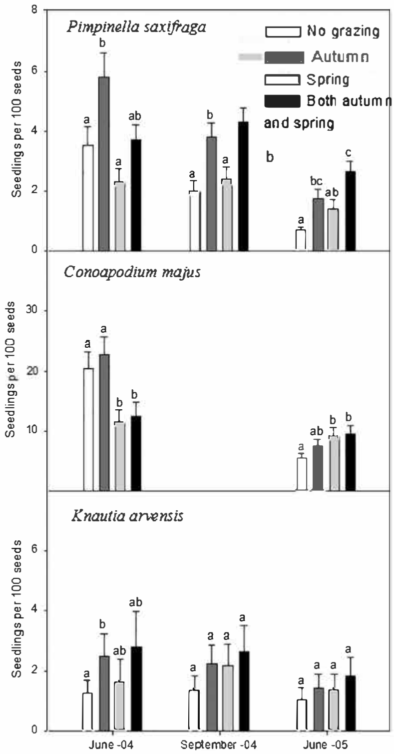

Figure 1. Seedlings per 100 seeds sown (mean ± standard error) in different grazing regimes in June and September 2004 and June 2005. Different letters indicate significant differences between grazing treatments within each census. Note the different scales for the response.

Conclusions

The effect of grazing depended on multifaceted relationships between the timing of grazing (spring or autumn, or both), plant species and the developmental stage of the seedlings. The results from this study suggest that spring and autumn grazing should be considered as a part of management in species-rich hay meadows. In particular, autumn grazing can be important in ecological restoration, as it minimizes litter accumulation and creates gaps in the sward that facilitate seedling establishment. Although spring grazing had a positive effect on final establishment, adverse effects in some species have been reported by others (Grime 2001) and spring grazing should therefore be used with some caution.

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The impact of soil-protecting technologies on soil erosion with maize sown on arable land and grassland

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Abstract

Two experiments were carried out at the HZS Jevíčko PLC site. The first experiment was established in May 2013 when plots were established by sowing maize into cover crops (woolly blue curls, crambe, rye) that had been sown in the autumn of 2012. Control plots without cover crop and fallow land were also included in the experiment. The inclination of the plot was 4.5°. The sowing was carried out with two types of seeding drills (Kinze 3000 and a prototype for strip-seeding PS-2) along the fall line. The second experiment was established in May 2015 when maize was sown into cultivated permanent grassland (Striger and Kuhn seeding drill) in a site located above a reservoir impounding water from the Želivka river constituting the main source of drinking water for Prague. The land inclination was 5-7°. In both experiments, soil erosion was measured by use of a field rainfall simulator. The highest soil loss measured after torrential rain (0.447 Mg ha^{-1}) was recorded on the fallow plot consisting of bare soil without vegetation. The rye plot (0.067 Mg ha^{-1}), the crambe plot (0.182 Mg ha^{-1}) and woolly blue curls (0.208 Mg ha^{-1}) had lower soil loss. Maize established into permanent grassland gave excellent soil protection with low soil erosion.

Keywords: erosion, soil protection technologies, cover crop, grassland, maize

Introduction

Out of the area of arable land (3.5 million ha) in the Czech Republic about 31% is threatened by water erosion and nearly 9% by wind erosion of soil. Accelerated erosion on farming land seriously threatens important functions of soil. Soil degradation by water erosion is a significant factor decreasing both productive and non-productive soil potential. Water erosion signs are closely linked to the quality of the soil given by its characteristics (physical, chemical and biological), and also by its utilisation, agronomical practices and rate of degradation (Janeček, 2012). The total number of biogas stations in the Czech Republic has increased from 13 to 692 in the last 10 years and their orientation on maize as the most effective energy plant brings a lot of threats, especially soil erosion (Usťak *et al.*, 2013).

Materials and methods

Two experiments were carried out at HZS Jevíčko PLC site in the Czech Republic (average annual temperature 7.4 °C, annual long-term rainfall average 545 mm, altitude 342 m, geographic coordinates: 49°37'43", 16°43'54"). The first experiment was established in May 2013 by sowing maize (*Zea mays* L.) into cover crops (woolly blue curls (*Phacelia tanacetifolia*), crambe (*Crambe abyssinica* (L.) Hochst.), rye (*Secale cereale*)) that had been sown in the autumn of 2012. Control plots without cover crop and fallow land were also included in the experiments. The inclination of the plot was 4.5°. The sowing was carried out with two seeding drills (Kinze 3000 and a prototype for strip-seeding PS-2) along the fall line. The second experiment was established in May 2015 by sowing maize into a cultivated strip in permanent grassland (with Striger and Kuhn seeding drill) in the site located above the reservoir Želivka which is the main source of drinking water for Prague. The slope inclination was 5-7°. Permanent grassland was treated by glyphosate herbicide at a rate of 4 l ha^{-1} before seeding.

In June 2013 and 2015, at the height of maize stand of 1 m, measurement of erosion prevention efficiency was carried out using a field rainfall simulator made by RISWC Prague in both of these experiments (Table 1).

Results and discussion

The highest soil loss measured with the field rain simulator after torrential rain (0.447 Mg ha^{-1}) was recorded on the fallow treatment consisting of bare soil without vegetation, whereas the lowest erosion was recorded on the rye (0.067 Mg ha^{-1}), followed by the crambe (0.182 Mg ha^{-1}) and woolly blue curls (0.208 Mg ha^{-1}) plots. Sowing technologies were even (Table 2), the loss of soil on the Kinze 3000 plots was 0.150 Mg ha^{-1}, on the PS-2 plots it was 0.154 Mg ha^{-1}. Average over cover crops, the soil loss was 0.152 Mg ha^{-1} that is about 1/3 compared to the soil loss measured for the fallow treatment. This demonstrates a significant erosion prevention effect of cover crops in maize grown to the height of about 0.9 m. The experiment also demonstrated significant effects of vegetation cover in comparison with fallow land (bare soil without vegetation) with respect to prevention of soil loss. Winter rye showed higher erosion prevention potential than woolly blue curl and crambe because it left more after-harvest residues in the spring.

The largest soil lost, also in the first location, occurred on the plot with the variant of fallow land (1.410 Mg ha^{-1}) (Table 3). The recorded soil loss was significantly lower on the plot with the experimental variant that was cultivated in the strips by using the Striger machine for cultivation and Kuhn for sowing

Table 1. Methodology of measurement with the field rainfall simulator:

Number of modes	1
Jet nozzle	30WSQ
Pressure [bar]	0.50
Time [min]	20
Valve	V2 + V3 together
Total precipitation [mm]	20.48
Intensity of precipitation [mm.min^{-1}]	1.02

Table 2. The results of measurements of soil loss (Mg ha^{-1}) with soil protection technologies of sowing into cover catch crops.

Alternative cover crops	Soil protection technologies		Average
	Kinze 3000	PS-2	
Wooly blue curls	0.212	0.204	0.208
Crambe abyssinica	0.180	0.184	0.182
Rye	0.059	0.074	0.067
Average of cover crops	0.150	0.154	0.152
Control (maize sown without cover crop)	0.432	0.392	0.412
Fallow land (bare soil cultivated 3 days before measurement with a rotavator)[1]			0.447

[1] Fallow land, only one measurement.

Table 3. The results of measurements of soil loss (Mg ha^{-1}) with soil protection technology Striger and Kuhn of sowing maize into grassland.

	Total soil loss (Mg ha^{-1})
Striger and Kuhn seeding drill	0.220
Fallow land (bare soil cultivated 3 days before measurement with a rotavator)	1.410

(0.220 Mg ha^{-1}). In 2015, the total dry matter production of maize sown into the permanent grassland reached 10.5 Mg ha^{-1} in the second experiment.

Conclusions

Increased demand of maize for biogas production brings a lot of threats to agricultural land, increased risk of soil erosion being a major threat. The experiments demonstrated significant preventive effects of vegetation cover (maize) on soil loss in comparison with fallow land. Growing maize with cover crops resulted in lower erosion than did growing maize alone. Development of new soil protection technologies brings new possibilities for soil erosion decrease.

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Theme 5.
Grassland in a changing climate – perspectives on mitigation and adaption

Role of European grasslands in the mitigation of climate change – potential, constraints and research challenges

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Abstract

There is a growing consensus that projected levels of meat demand are unsustainable, so we will need to eat less meat in the future, as well as tackling waste in the food system. Since ruminants produce large quantities of methane through enteric fermentation, it has been suggested that ruminants should be the primary target for livestock product reduction, but there is a strong case to instead target reductions in monogastric livestock production, since monogastrics consume potentially human-edible products, whereas ruminants are able to consume products (grass) that cannot be consumed by humans, and are the most efficient means of producing human food on land unsuitable for crops. The livestock sector and grasslands play a major role in climate change, from both global and European perspectives. This is illustrated by data on the current status of emissions from the livestock sector in combination with projections of emissions from the sector to 2050. There is, however, also a large climate change mitigation potential in the livestock/grassland sector, both via supply-side measures and demand-side measures. Here we provide evidence from the literature showing that the grazing lands that support ruminant production in Europe are important for carbon and nitrogen cycling, and for underpinning a range of other ecosystem services, adding further weight to the case to retain pasture raised ruminants as a part of future food systems. This will need to be done, however, in the context of reduced overall consumption of livestock products. Indeed, reduced livestock product consumption could be seen as the enabling condition that allows pasture-fed ruminants to take their role in future food production systems. If more of human dietary requirements were met through vegetal matter, which is more efficient in terms of land, greenhouse gases, water and other resources, there is more 'headroom' in the food system to consider a range of production systems to deliver livestock products to human diets. Pasture-fed ruminants, with high environmental and welfare status, could play a central role in future food systems, in delivering food security and in protecting the ecosystem services provided by grasslands.

Keywords: climate change, ruminants, supply-side measures, demand-side measures, emissions

Introduction

Globally, the livestock sector constitutes twenty billion animals (FAOSTAT, 2016), and it provides direct livelihood and economic benefits to at least 1.3 billion producers and retailers (Thornton, 2010; Herrero *et al.*, 2009). As an economic activity, livestock contributes to 40-50% of agricultural GDP globally (FAO, 2009).

Livestock have a significant environmental footprint, occupying 30% of the terrestrial land area for grazing, using 32% of global freshwater resources (FAO, 2006), and with one-third of the global cropland area devoted to producing animal feed (FAO, 2006).

In this paper we assess the role of the livestock sector and grassland in the mitigation of climate change, from both global and European perspectives. We begin by assessing the current status of emissions from the livestock sector, then summarise projections of emissions from the sector to 2050. We then examine the technical and economic climate mitigation potential available in the livestock sector, first by focussing on supply-side measures (feeds, breeding, dietary additives, system level approaches, large vs small scale production in developed vs developing countries), and then on demand-side measures (e.g. diet, waste). Finally, we examine the role of grasslands in ecosystem services such as carbon (C) and nitrogen (N) cycling, before synthesising all of this information to consider the role of pasture fed ruminants in the sustainable food future, and research needs to deliver this.

Current status of emissions from the livestock sector

Recently, Herrero *et al.* (2016) collated estimates (EC-JRC/PBL, 2011; US-EPA, 2006; Popp *et al.*, 2010; Tubiello *et al.*, 2013; Herrero *et al.*, 2013; FAO, 2010; Gerber *et al.*, 2013) of total greenhouse gas emissions from livestock for the period 1995-2005. Total livestock emission estimates were between 5.6 and 7.5 Gt CO_2eq yr^{-1}. The most important contributors to this total were enteric methane (1.6-2.7 Gt CO_2eq yr^{-1}), N_2O emissions associated with feed production (1.3-2.0 Gt CO_2eq yr^{-1}) and land use for animal feed and pastures, including land use change (~1.6 Gt CO_2eq yr^{-1}; Herrero *et al.*, 2016).

Cattle production dominates emissions from the livestock sector (64-78% from Herrero *et al.*, 2013; Gerber *et al.*, 2013; FAO, 2013). Using Life Cycle Assessment, FAO (2013, 2014) estimated cattle emissions from all sources to be about 4.6 Gt CO_2eq yr^{-1}, of which 2.5 Gt CO_2eq yr^{-1} were from beef cattle and 2.1 Gt CO_2eq yr^{-1} from dairy cattle (producing both milk and meat). Other species have much lower and similar levels of emissions: buffalo (0.6 Gt CO_2eq yr^{-1}) and small ruminants (0.5 Gt CO_2eq yr^{-1}) and other livestock that do not use grassland directly: poultry (0.7 Gt CO_2eq yr^{-1}), pig (0.7 Gt CO_2eq yr^{-1}; Herrero *et al.*, 2016).

Total greenhouse gas (GHG) emissions from livestock farming in the EU-27 are estimated to have been about 0.5 Gt CO_2eq yr^{-1} in the last decade and are slowly decreasing (Leip *et al.*, 2010; EEA, 2015).

Projections of future trends in emissions from the livestock sector

Global per-capita consumption of livestock products has more than doubled since 1970 (FAO, 2009). Increasing global population, higher incomes, and urbanization are projected to drive increases in the consumption of milk and meat over the next twenty years, at least at previously observed rates (Rosegrant *et al.*, 2009), with most of the growth projected to occur in developing countries. In response to these demand trends, production has increased significantly. Beef and milk production have more than doubled since 1970, and the production of monogastrics (pigs and poultry) has grown by a factor of five or more in some regions (Thornton, 2010).

Intensification of production, in terms of increased livestock and/or crop productivity, has been central in raising output per unit of land and animal (FAO, 2006). But while intensification has occurred in some regions, agricultural land expansion has also been an important component of production growth in some regions, such as Africa and Latin America (Herrero *et al.*, 2016). These trends, if continued, could drive significant increases in GHG emissions, deforestation, loss of biodiversity, and other negative impacts on the environment (Smith *et al.*, 2013).

Estimates of emissions associated with the projected growth of the livestock sector to 2050, reported in Herrero *et al.* (2016) suggest that methane from enteric fermentation, methane from manure management, and nitrous oxide from manure management are likely to grow at rates between 0.9-5%, 0.9-4%, 1.2-3% per year, respectively (US-EPA, 2006; Popp *et al.*, 2010; Bodirsky *et al.*, 2012; US-EPA, 2013; Westhoek *et al.*, 2011; Havlik *et al.*, 2013). The ranges reflect different scenarios and assumptions about growth in demand for livestock products, animal numbers and type, and productivity growth in livestock systems. A continuation of existing trends would lead to increases in livestock emissions of between 1-1.5% per year across all sources other than land-use change (US-EPA, 2006; Havlik *et al.*, 2013; reported in Hererro *et al.*, 2016). Although not only attributable to livestock, emissions from deforestation over the same period are projected to grow at a slower rate of 0.8% per year (Havlik *et al.*, 2013). Cropland area expansion is growing at a faster rate than pasture expansion, primarily due to the accelerated growth of pig and poultry production (>5% globally – though these do not use grasslands directly; Herrero *et al.*, 2016).

According to Gerber and Mottet (2015) the European livestock sector could curb its direct GHG emission intensity by about 15 to 20% between 2005 and 2025 and achieve a 30 to 50% reduction by 2050. Efficiency gains in other regions will probably be higher than in the European Union, since the majority of European production systems are already relatively efficient.

Climate change mitigation potential in the livestock/grassland sector; supply-side measures

Technical options for GHG mitigation in the livestock sector include animal-based options, which can be categorized as targeting enteric methane, manure storage and application or deposition, animal management, and grazing land management, which includes measures such as improved management regimes, fire management and improved nutrient management (Smith *et al.*, 2008; Herrero *et al.*, 2016). In ruminant production systems, enteric methane emissions usually comprise the largest proportion of GHG emissions and have been the main focus for animal-based mitigation research (Ripple *et al.*, 2014). Animal-based options such as feed additives (including ionophores, probiotics, enzymes, extracts), feed with greater digestibility, and manure management are estimated to have a combined GHG mitigation potential of 0.01-0.5 Gt CO_2eq yr^{-1} (Herrero *et al.*, 2016).

Improved grazing land management practices can include reductions in methane emissions (wetland drainage) or nitrous oxide (e.g. through fertilizer management), though most operate through increasing soil carbon stocks. Practices that impact upon species composition, forage consumption, nutrient and water inputs, and fire can impact upon soil carbon stocks (Smith *et al.*, 2008; Herrero *et al.*, 2016). Changes in grazing management could lead to annual sequestration of up to 150 Mt CO_2eq yr^{-1} globally (Henderson *et al.*, 2015). About 81% of this potential is in developing countries, in areas where production is predicted to increase following a period of de-stocking. The use of legumes in pastures has been estimated to sequester 200 Mt CO_2eq globally, though this could increase soil N_2O emissions by 60 Mt CO_2eq yr^{-1}, offsetting 28% of the soil carbon sequestration benefits according to Henderson *et al.* (2015). About half of the global net mitigation potential of this option is in developing countries (Henderson *et al.*, 2015).

The metrics used to measure mitigation are also important. Reducing emissions per animal or per hectare may appear effective, but if productivity is greatly reduced, the GHG emissions per unit of agricultural product (i.e. the emissions intensity) may not be decreased (Bennetzen *et al.*, 2012; Bennetzen *et al.*, 2016). In this case, production will need to be increased elsewhere to meet the same demand, so emissions may be displaced elsewhere. For this reason, emissions intensity can be considered the best metric for GHG mitigation in agriculture.

Sustainable intensification (Smith, 2013), whereby productivity relative to GHG emissions is increased, can help to deliver reduced emissions intensity, with evidence that this has been occurring over recent decades (Smith *et al.*, 2014; Bennetzen *et al.*, 2016). Options in the livestock/grassland sector in this category include improved animal productivity and health, such as improved genetic potential of animals for production, and improved reproductive performance, health, and live-weight gain rates (Herrero *et al.*, 2016). An increased longevity of dairy cows increases emissions intensity since the effect of the first non-productive years is spread over a larger number of years. Herrero *et al.* (2016) estimated that improved animal management practices could reduce GHG emissions in the livestock sector by 0.2 Gt CO_2eq yr^{-1} by 2050. Increased grassland productivity could also reduce deforestation pressure (Oliveira Silva *et al.*, 2015). This could be done through high-tech solutions such as genetic improvement, or simpler options such as rebalancing the distribution of inputs to optimise production and close yield gaps (Mueller *et al.*, 2012).

Climate change mitigation potential in the livestock/grassland sector; demand-side measures

Food demand is expected to increase by 60-100% by 2050, due to increasing populations and per-capita wealth (Royal Society of London, 2009). Resource-use efficiency of livestock production is low compared to crop production, and around one third of the world's cereal production is fed to animals (FAO, 2006). Meat currently represents only 15% of the total energy in the global human diet, yet approximately 80% of agricultural land is used for animal grazing or the production of feed and fodder for animals (FAO, 2006), though this includes extensive grasslands in areas where other forms of agriculture would be extremely challenging. On average, the production of beef protein requires about 50 times more land than the production of vegetable proteins (Nijdam *et al.*, 2012), and greenhouse gas emissions excluding land-use change are about 100 times higher, so reducing consumption of livestock products could greatly reduce the need for more food and reduce the climate impact of food production.

The global adoption of healthy diets (following healthy eating recommendations; Stehfest *et al.*, 2009; Bajželj *et al.*, 2014; Tilman and Clark, 2014), adequate food production in 2050 could be achieved on less agricultural land than currently used, allowing regrowth of natural vegetation, and reduced greenhouse gas emissions by 4.3 Gt CO_2eq yr^{-1} compared with the baseline. More extreme scenarios, modelled by Stehfest *et al.* (2009) would yield emission reductions of 5.8, 6.4, and 7.8 Gt CO_2eq yr^{-1}, respectively, for a no ruminant meat, no meat, and no animal product scenario. In addition to reducing pressure on agricultural land, a global transition to a healthy, low-meat diet would reduce the mitigation costs, particularly in the energy sector, of achieving a 450 ppm CO_2eq stabilisation target in 2050 (Stehfest *et al.*, 2009). While there may be considerable barriers to implementation, reducing demand for livestock products has a large technical mitigation potential (Popp *et al.*, 2010; Smith *et al.*, 2013; Bajželj *et al.*, 2014; Tilman and Clark, 2014; Hedenus *et al.*, 2014; Westhoek *et al.*, 2014; Lamb *et al.*, 2016).

Reduced consumption also frees land for other uses, so called 'land sparing' (Lamb *et al.*, 2016), which can be used for bioenergy, carbon sequestration and/or biodiversity conservation (Erb *et al.*, 2012; Smith *et al.*, 2013). Switching to a low-animal product diet that converges to the global average energy demand in the year 2000 (i.e. 2,800 kcal cap^{-1} d^{-1}, compared with the global mean of 3,100 kcal cap^{-1} day^{-1} in the reference case) gave emission reductions of 0.7-7.3 Gt CO_2eq yr^{-1} if the 'spared land' is used for bioenergy, and 4.6 Gt CO_2eq yr^{-1} if afforestation is assumed (Smith *et al.*, 2013).

Considering plausible low-meat diets, Herrero *et al.* (2016) estimated a mitigation potential of 4.3-6.4 Gt CO_2eq yr^{-1}, with about 1-2 Gt CO_2eq yr^{-1} coming from process emissions, mostly CH_4 and N_2O, and the remainder from land-use change CO_2. Given that projected levels of livestock consumption to 2050 cannot be sustained, demand management must occur (e.g. reduced consumption of all livestock products in diets, and reduced food waste; Smith, 2014). Under these conditions, pasture-fed ruminants will have a role in future, sustainable food systems, since they convert non-human-edible material into human-edible food most effectively on land not suitable for crop production (Gill *et al.*, 2010).

Role of grasslands in carbon and nitrogen cycling

European grasslands are characterized by a huge variety of management practices and varying management intensity, ranging from high-intensity grasslands, mown up to seven times per year with high mineral fertilizer inputs (e.g. in places in Ireland and Scotland) to more extensive systems or even abandoned grasslands, such as those at higher altitudes in the Alps. The four most common grassland types in Europe are: (1) short duration grass leys; (2) legume based leys; (3) sown intensive grasslands and (4) permanent grasslands (based on results presented by Soussana *et al.,* 2004 for France, which are relevant for most of Europe). During the last 50 years, the European grassland area has significantly reduced, by 15 M ha in favour of the production of fodder maize and other annual crops (FAOSTAT, 2011).). This reduction was the result of intensification of grassland and animal production, decrease in cattle population, use of concentrates and soybean in the ration, abandonment, and the effect of EU-policy (Huyghe *et al.*, 2014). Even marginal grasslands tend to be abandoned, particularly in mountainous and Mediterranean areas, where they can be of crucial importance for preserving biodiversity, protecting soils against erosion and maintaining the local population density. The observed reduction differs between countries. Losses were high in Belgium, France, Italy and the Netherlands while the grassland area remained almost stable in Luxembourg and the United Kingdom. Nevertheless, grasslands still cover the largest proportion of the agricultural area. In 2007, permanent grasslands covered over 57 million ha in the EU-27 and temporary grasslands about 10 million ha, which represents 33% and 6%, respectively, of the total utilised agricultural area in the EU-27.

Besides essential management practices, which involve the removal of biomass and subsequently C and N from the system via harvests and/or grazing, other activities further affect carbon C and N cycling in grassland ecosystems. These include site amendments in the form of mineral or organic fertilizer to compensate C and N removals, and residue incorporation to ensure productivity at a constant level. Further management practices include over-sowing, occasional harrowing and rolling and pesticide/herbicide applications to maintain the favoured vegetation composition. Less frequently restoration, particularly of permanent grasslands, may become necessary in the form of ploughing (Merbold *et al.*, 2014). Similar activities occur during land conversion from cropland to grassland and *vice versa*.

In addition to the multitude of management practices, grassland systems differ in species composition. While some are single-species ecosystems – pure monocultures of *Lolium perenne* L. (e.g. often found in Ireland and Scotland) – others are more diverse and based on multispecies mixtures and often include legumes (e.g. Central European grasslands). This complexity in grassland ecosystems has substantial consequences for biogeochemical cycling of nutrients, primarily of C and N, that are not yet fully understood. However, detailed knowledge on the advantages and disadvantages of specific activities, as well as internal processes, is needed since grasslands have been highlighted as valuable systems to mitigate climate change (Henderson *et al.*, 2011; Schulze *et al.*, 2009; Freibauer *et al.*, 2005; Smith *et al.*, 2005).

Carbon cycling in grasslands

The mitigation potential of grasslands has often been associated with the potential for soil carbon sequestration, a process where C from the atmosphere is 'locked away' in the soil (Powlson *et al.*, 2011; Lal, 2004; Conant *et al.*, 2001; Soussana *et al.*, 2004). The primary processes and their net effects determining carbon sequestration are photosynthesis and respiration and the balance of these leads either to soil carbon storage or soil carbon loss. Photosynthesis – the process binding CO_2 in plant tissue from the atmosphere – is largely driven by environmental conditions (radiation, temperature and water availability) as well as biological pre-requisites including soil fertility, biodiversity and anthropogenic disturbance. In addition, a reduction in aboveground vegetation by mowing leads to a decline in photosynthesis and autotrophic respiration.

Soil respiration, comprising mycorrhizal, rhizosphere and soil basal respiration (Moyano *et al.*, 2007), is driven by a similar set of driver variables with photosynthesis having a profound influence on respiratory losses by regulating root activity (Xu and Baldocchi, 2004). Harvest and grazing can additionally lead to an increase in respiration losses, due to reduced soil cover affecting soil temperature, while at the same time reducing soil water content due to increased evaporative water losses. It is important that these counteracting effects are precisely understood. The interaction between the various processes differs from ecosystem to ecosystem and quantification remains challenging (Gilmanov *et al.*, 2007). Overall, the two individual processes photosynthesis and respiration have been studied individually and jointly and are well constrained (e.g. Law *et al.*, 2002; Gilmanov *et al.*, 2007; Schulze, 2006). Specific grassland management activities and an intensification of grassland systems, either by increasing the stocking density or by changing fertilizer management, and a subsequent increase in harvest can lead to accelerated C and N turnover within the system and subsequently to changed stocks of soil carbon (Powlson *et al.*, 2011; Soussana *et al.*, 2007).

While harvest leads to a direct export of C, some of this C might be returned in form of organic manure. Therefore, the effects of grazing are multidimensional and depend largely on grazing intensity (Soussana *et al.*, 2010; Freibauer *et al.*, 2004; Smith *et al.*, 2008). Under intensive grazing between 25-40% of the C that was taken in by animals is returned to the soil in form of faeces, leading to a direct recycling of C. However faeces may decompose and C will be lost in form of CO_2 to the atmosphere (Hopkins *et al.*, 2009), while the remaining C is relocated into the soil. Furthermore, C is incorporated into the animals' meat (low percentage) or exhaled in form of CH_4 caused by rumination (Johnson and Johnson, 1995). A quantification of these entire C flows in the field remains challenging. Stocking rate is a crucial factor influencing carbon cycling with the two factors identified working in opposite directions. While grazing intensity and the associated C returns in form of faeces could favour net primary production (De Mazancourt *et al.,* 1998), plant intake by the animals leads to a reduction in leaf area index and subsequent decline in CO_2 capture via photosynthesis.

Grazing has become less popular in Europe in recent decades as can be seen from inventories of the (EGF Working Group 'Grazing'; Van den Pol-Van Dasselaar *et al.*, 2015). Trends that caused a decline in the popularity of grazing include the management of larger herds and introduction of automated milking systems, land fragmentation, but also factors such as weather and difficulties with grassland management (timing of grazing/cutting, fertilisation). These factors are relevant for farmers who have to manage the quantity and quality of their grassland production.

Enhanced carbon storage is not only important for climate change mitigation but certainly has additional benefits, including better soil quality and functioning (Powlson *et al.*, 2009). Both properties support vegetation cover and soil organic matter and lead to additional benefits, such as improved water infiltration of the soil and a reduced likelihood of erosion (Blair *et al.*, 2006; Watts *et al.*, 2006). At the same time, management activities that improve soil carbon content may have detrimental effects, as recently summarized by Powlson *et al.* (2011). Noteworthy among these are enhanced GHG emissions in the form of N_2O, which can easily outweigh small gains in soil organic C. Additional critical issues remain when aiming at increasing carbon sequestration in grasslands to mitigate climate change. A critical limitation is that soil C storage is finite and will reach equilibrium over time (Johnston *et al.*, 2009; Smith, 2014). Furthermore, once a new, higher equilibrium soil C level is reached, the land management responsible for this increase needs to continue since the process is reversible (Freibauer *et al.,* 2004; Smith, 2014). These well studied issues lead to the conclusion that soil C sequestration in grasslands is only one part of the argument, and that other mitigation activities are necessary.

Nitrogen cycling in grasslands

Nitrogen cycling in grasslands is slightly more complex than carbon cycling. This is mainly due to a larger variety of ecosystem processes that drive nitrogen turnover, as well as by the fact that carbon is available in the atmosphere and can be accessed by plants directly. Nitrogen in contrast, even though it is abundant (78% in the atmosphere) is only available in inorganic molecular form, and transformation to organic plant accessible form is strongly energy dependent (Galloway, 1998). Because of this, ecosystems in general are characterized by large internal N re-cycling (Rennenberg *et al.*, 2009). Similarly to C, N cycling in grasslands largely depends on environmental conditions including climate and soil properties (Butterbach-Bahl *et al.*, 2011). Management activities play a crucial role in ecosystem N cycling, either by removing large amounts of N during harvest, or by supplying considerable amounts of N in form of amendments (mineral and organic fertilizers), or even indirectly via atmospheric N deposition (Galloway *et al.*, 2008). Even though the productivity of an ecosystem largely depends on the efficiency of N cycling, several pathways that lead to N losses have been identified. The two most important losses occur either via leaching of N in form of NH_4^+, NO_3^- and DON (Tilman *et al.*, 1996) and/or in gaseous form due to volatilization (NH_3 and NO_x) or as a by-product of soil microbial processes (in the form of N_2O, N_2), which were reviewed by Butterbach-Bahl *et al.* (2013) recently. All losses lead to additional detrimental effects, such as ground water pollution (Di and Cameron, 2002) and/or greenhouse gas emissions (Stehfest *et al.*, 2006). These losses are particularly favoured when N is available in the soil in excess compared to the N needs of plants (Ledgard *et al.*, 1999; Ball *et al.*, 1986). Additional N turnover and N losses occur due to grazing activities. N is incorporated into the animal's body by feeding on forage while at the same time large amounts of N are returned to the system in form of urine and faeces. Urine patches in particular have been shown to lead to considerable losses of N in the form of N_2O (Oenema *et al.*, 1997; Butterbach-Bahl *et al.*, 2013).

Again management activities are key to optimizing N cycling in grasslands. Appropriate timing and the overall amount of N added to a system defines the amounts of N lost from the system. Tailoring the N inputs to the plant demand under favourable environmental conditions is therefore essential (Butterbach-Bahl *et al.*, 2011). N inputs can also be delivered by legumes with their associated rhizobia via biological nitrogen fixation (BNF). These have been used to reduce the climate footprint of grassland, by reducing the application of mineral fertilizers and at the same time delivering better nutritional value of the products from the system. Figure 1 shows major carbon and nitrogen flows in European grassland ecosystems.

Potential solutions to close the cycles of C and N in grasslands and research needs

Each of the processes highlighted in the previous sections lead to changes in C and N in grassland ecosystems. However the interactions between C and N cycling are manifold. This is particularly important, as it implicates that sustainable intensification of grassland ecosystems can only be achieved with a holistic approach, including all element cycles, C and N as well as phosphorous, sulphur and micronutrients.

A great potential to improve C and N cycling and mitigate GHG emission in grasslands is suggested by replacing conventional fertilizer application by BNF via multi-species grasslands (Luescher *et al.*, 2014). Such management change has to be implemented wisely since increasing the legume content above a certain threshold does not lead to infinite N fixation (Nyfeler *et al.*, 2009). At the same time, implementing legumes in single species grassland or in organic soils, may be challenging given the amount of energy needed to biologically fix nitrogen. This energy need may be a negative criterion for species competition in already intensively used swards. In contrast, once legume establishment is successful, legumes can substantially contribute to the improvement of nutrient poor grasslands (Suter *et al.*, 2015;

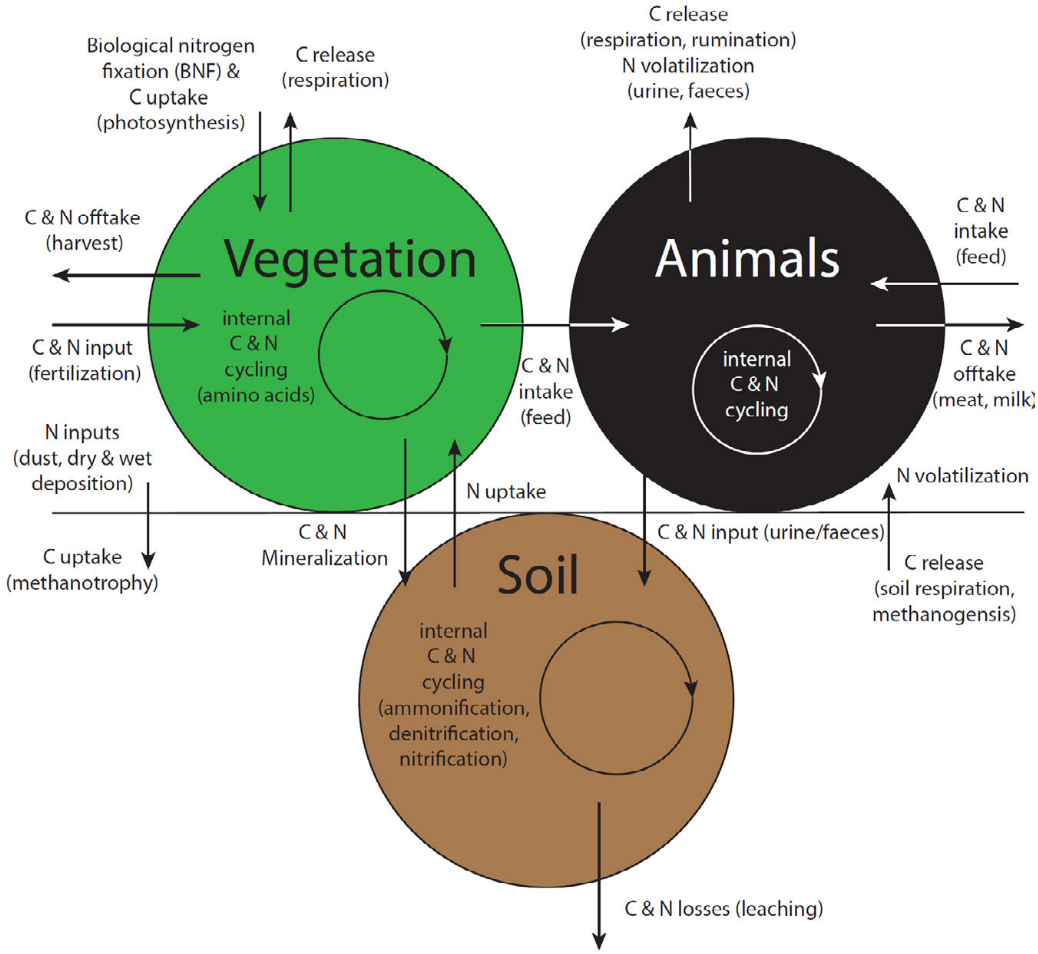

Figure 1. Major carbon (C) and nitrogen (N) flows in European grassland ecosystems. Numbers differ across systems depending on the management intensity, and whether the grassland is primarily used for feed production (e.g. hay, silage) or grazed.

Soussana *et al.*, 2010). Additional positive effects such as better digestibility, and consequently reduced CH_4 losses via enteric fermentation, have been identified (Soussana *et al.*, 2010). Lüscher *et al.* (2013) summarised the overall potential of legume-based grassland-livestock systems in Europe and concluded that forage legumes offer important opportunities for tackling future agricultural challenges. They showed that the potential of legumes for sustainable intensification is related not just to one specific feature; the strength of legumes stems from the fact that several of the legume features can act together on different 'sites' in the soil-plant-animal-atmosphere system. The advantages of legumes are most effective in mixed swards with a legume abundance of 30-50% (Lüscher *et al.*, 2013). These multiple advantages benefit the whole grassland-husbandry system through reduced dependency on fossil energy and industrial N fertilizer, lower nitrate and greenhouse gas emissions into the environment, lower production costs, higher productivity and protein self-sufficiency.

Other management options to mitigate climate change and similarly leading to a sustainable intensification in grasslands include: (1) a system specific definition of the optimal grazing intensity as suggested by Lemaire (2012); (2) reduced tillage (Six *et al.*, 2004); (3) better C:N coupling (Soussana and Lemaire, 2014); (4) pasture restoration (Conant *et al.*, 2001); (5) overall improved forage quality to favour digestibility and subsequent C turnover amongst few others (Soussana and Lemaire, 2014); (6) improved genetics/plant breeding which can lead to improved crop varieties and/or types (e.g. early maturing, drought resistant varieties or crop types) which are resilient against drought or disease – thereby increasing crop yields and reducing yield variability (Bryan *et al.*, 2011).

Potential management options need to be defined individually for the respective grassland ecosystems. In other words, a degraded grassland has to be treated differently to a high-intensity dairy production system, and differently from an extensive alpine meadow. In order to identify the best practice for a specific system, multi-disciplinary research approaches are key. Such approaches should not only focus on nutrient gains and losses, but include socio-economic and environmental developments. The adoption of management activities by farmers will depend on factors such as productivity impacts, costs, future vs ready-to-use measures, applicability by farmers, acceptability for farmers (Van den Pol-Van Dasselaar and Bannink, 2014). Ultimately, farmers will only adopt management activities if the output still allows profitable production (Finger *et al.*, 2010). Moran *et al.* (2011) suggested three basic criteria to be used to guide research on mitigation and adaptation measures: (1) technical effectiveness (which helps to define the measures that actually work in a variety of farm environments); (2) economic efficiency (preferably cost-beneficial) and (3) equity (of the impacts of measures).

Similarly, population increases and changes in dietary preferences need to be considered, as these may lead to additional pressure on currently relevant land management systems. Besides the system-wide approach, additional research is needed on currently available management adjustments in terms of the resilience of grassland systems to environmental change (Lüscher *et al.*, 2005; Soussana and Lüscher, 2007), comprising not only overall higher temperatures and changed precipitation patterns in the near future (IPCC, 2013, Ciais *et al.,* 2013), but also the effects of extreme climatic events (Ciais *et al.,* 2005; Reichstein *et al.*, 2013). Following the suggestions provided by Kayler *et al.* (2015) experiments are needed that go beyond currently tested climatic thresholds.

Role of grasslands for providing other ecosystem services

European grasslands have not only a role in the mitigation of climate change and in C and N cycling, but they provide many more ecosystem services. Grasslands serve multifunctional purposes ranging from provision of autochthonous fodder for animal husbandry (and hence food-provision for citizens) to biodiversity, to the provision of traditional originated landscapes that European citizens appreciate for recreational purposes (Van den Pol-Van Dasselaar *et al.*, 2014).

In general the economic functions of grasslands, mainly feed for herbivores, are considered the most important. It is expected that this will remain unchanged in the future. The economic functions apart from providing feed for herbivores for the production of meat, milk or wool, also include feed for bees (honey production) and, more recently, economic functions such as providing plant fibre and providing raw material for bio-energy production. Environmental functions of grasslands are generally placed second. These functions include adaptation to climate change and mitigation of climate change, especially via carbon sequestration, although forestry is seen as a strong competitor for this function. This, however, is still debatable, and more research seems to be necessary on the potential of grasslands compared with forestry or arable cropping for climate mitigation. Grasslands also affect water quality and quantity. The effect depends on the region in Europe: in some regions sufficient supply and protecting soil from erosion is important, in other regions providing flood plains is relevant. Another important function of grasslands is biodiversity. In many European areas grasslands provide unique ecosystems. Preserving these ecosystems adds to the global challenge to maintain biodiversity.

Grasslands also deliver social services. Apart from providing livelihood (lifestyle, labour, income; mentioned under economic functions), grasslands and their management are often part of the local culture as well. In some countries like the Netherlands, grazing clearly contributes to the image of dairy farming. Amenities such as landscape for tourism and space for recreation and sports are provided by grasslands. Grasslands contribute to maintaining populations in marginal areas.

The Millennium Ecosystem Assessment report (MEA, 2005) distinguished four groups of ecosystem services: (1) provisioning services: products obtained from ecosystems, e.g. production of food, water; (2) regulating services: benefits obtained from the regulation of ecosystem processes, e.g. control of climate and disease; (3) cultural services: non-material benefits people obtain from ecosystems through spiritual enrichment, cognitive development, reflection, recreation, and aesthetic experiences, e.g. recreation and beauty of the landscape; and (4) supporting services: ecosystem services that are necessary for the production of all other ecosystem services, e.g. nutrient cycles, crop pollination. Grasslands contribute to all these four groups. The importance of grasslands for stakeholders in Europe was studied by Van den Pol-Van Dasselaar *et al.* (2014) using questionnaires amongst stakeholders like primary producers, policy makers, researchers, advisors, NGO's, industry and education. When asked about the importance of different aspects of sustainability, stakeholders, on average, valued economic aspects the highest, followed by ecological aspects and finally social aspects. There were, however, differences between countries and stakeholder types. The results of the questionnaires also showed that many individual functions of grasslands are highly recognized and appreciated by all relevant stakeholder groups (Table 1).

Grasslands, with their multifunctional roles, can provide a good basis for developing sustainable production systems in the long term. The increasing global demand for meat and milk, environmental concerns about the sustainability of intensive production systems, and concerns about food quality and safety favour an increasing role for grassland-based ruminant production systems in the future. Furthermore, grassland is the only land use option which is capable of delivering a large number of ecosystem services simultaneously.

Is there a role for pasture-fed ruminants in a sustainable food future?

There is a growing consensus that projected levels of meat demand are unsustainable, so we will need to eat less meat in the future (Bajželj *et al.*, 2014; Tilman and Clark, 2014; Hedenus *et al.*, 2014; Westhoek *et al.*, 2014), as well as tackling waste in the food system (Bajželj *et al.*, 2014). Since ruminants produce large quantities of methane through enteric fermentation, it has been suggested that ruminants should be the primary target for livestock product reduction (Ripple *et al.*, 2014), but there is a strong case to instead target reductions in monogastric livestock production (Schader *et al.*, 2015), since monogastrics consume potentially human-edible products, whereas ruminants are able to consume products (grass) that cannot be consumed by humans (Schader *et al.*, 2015; Herrero *et al.*, 2016), and are the most efficient means of producing human food on land unsuitable for crops (Gill *et al.*, 2014; Herrero *et al.*, 2016).

We have provided evidence that the grazing lands that support ruminant production in Europe are important for carbon and nitrogen cycling, and for underpinning a range of other ecosystem services, adding further weight to the case to retain pasture raised ruminants as a part of future food systems. This will need to be done, however, in the context of reduced overall consumption of livestock products (Hedenus *et al.*, 2014; Westhoek *et al.*, 2014). If climate impacts were considered economically (Wirsenius *et al.*, 2010), meat and livestock products would become more expensive, and this would lead to a situation where humans 'eat less and eat better' in terms of livestock product consumption. Indeed, reduced livestock product consumption could be seen as the enabling condition that allows pasture-fed ruminants to take their role in future food production systems. If more of human dietary

Table 1. Importance of functions of grasslands according to the respondents of a European questionnaire (1 = not important; 5 = very important) (n=1,798). Functions with an average score of more than 3 are given (Van den Pol-Van Dasselaar *et al.*, 2014).

Provisioning services	
High quality forage	4.1
Dairy cow milk production	4.0
Low cost animal feed	4.0
Nutritional quality of animal products for human consumption	3.9
Beef meat production	3.7
Global food production	3.5
Region of origin of animal products	3.5
Regulating services	
Biodiversity	3.8
Conservation of the quality of ecosystems	3.8
Water catchment	3.5
Erosion control	3.3
Carbon sequestration	3.3
Mitigating greenhouse gas emissions	3.3
Adaptation to climate change	3.2
Cultural services	
Beauty of the landscape	4.0
Positive perception of animal production systems	3.9
Rural development	3.4
Maintaining populations in rural areas	3.4
Cultural values	3.3
Tourism/recreation	3.3
Supporting services	
Grazing	4.2
Animal health	3.9
Animal welfare	3.9
Conservation of soil structure and fertility in cropping systems	3.8
Feed protein supply at farm level	3.7
Competitiveness of farming systems	3.5
N fixation via legumes	3.4
Availability of water	3.4

requirements were met through vegetal matter, which is more efficient in terms of land, greenhouse gases, water and other resources, then there would be more 'headroom' in the food system to consider a range of production systems to deliver livestock products to human diets. Under such a future, pasture-fed ruminants, with high environmental and welfare status, could play a central role in delivering food security and in protecting the important functions and ecosystem services provided by grassland.

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Climate challenges and opportunities in northern and southern Europe – role of management and exploitation of plant traits in the adaptation of grasslands

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Abstract

Climate change and its effects on grasslands, varies across Europe, with the Mediterranean and Nordic regions at the extreme ends. While productivity in the Mediterranean region will become even more limited by drought than today, productivity in the Nordic region may benefit from climate change, although there are several uncertainties regarding the possibility to utilize a longer thermal growing season, such as the interactions with Nordic light conditions and the effects of excessive precipitation. In both regions, climate change will lead to modifications of the annual productivity cycles, which will require adaptations in harvest and grazing regimes. The identity of species and mixtures with optimal performance is likely to shift somewhat in response to altered climate and management systems. Grasslands containing legumes can potentially utilize the fertilizing effect of elevated CO_2 better than many other crops or ecosystems, because the maintenance of sink strength by recurrent defoliation, and N provided by N fixation, limits photosynthetic acclimation to elevated CO_2. Breeding efforts of grassland species should be directed both towards improving plant strategies to cope with abiotic stresses, and towards adaptation to new seasonal patterns and managements, while taking care of possible trade-offs with productivity.

Keywords: climate change, drought, forage, Mediterranean, Nordic, plant breeding, winter survival

Introduction

Climate change poses significant challenges for grassland-based food production and other ecosystem services that these grasslands provide. Climate change may also imply some opportunities. The observed and projected climate change in Europe was summarized by Kovats *et al.*, (2014). The average temperature over land area during 2002-2011 was 1.3 °C above the 1850-1899 average, with substantial differences between regions and seasons. Warming has been strongest in Scandinavia, especially in winter, whereas the Iberian Peninsula has warmed mostly in summer. Warming is expected to continue, and be greater in winter in northern Europe and in summer in southern Europe. Precipitation has increased in northern Europe and decreased in southern Europe, a trend which is expected to continue. Extreme heat waves and droughts (mostly in the south), and heavy rains (mostly in the north), are also expected to increase in frequency. The response of grasslands to these changes in climate is complex. In addition to the direct effects of CO_2 concentration, temperature, precipitation and their interactions, there are important complicating factors such as plant competition and other plant-plant interactions, perennial growth habits, seasonal productivity patterns, and plant-animal interactions (Porter *et al.*, 2014). In this paper, we will contrast the climate challenges and opportunities for grassland production in northern and southern Europe. We focus on the Nordic region and areas with a Mediterranean climate, and explore the role of management and exploitation of plant traits for adaptation of grassland production to future climates. We consider challenges and opportunities in terms of productivity and forage quality, and will not take mitigation strategies into account.

Climate challenges and opportunities for grasslands in Mediterranean Europe

In the Mediterranean region, permanent grasslands constitute a much larger area than temporary grasslands (Huyghe *et al.,* 2014). Much of this area is semi-natural grassland dominated by annual species maintained by seed banks (Cosentino *et al.,* 2014). The species composition varies greatly between years, depending on previous seed production, dormancy and germination, which are processes strongly influenced by climatic conditions such as temperature and moisture (Cosentino *et al.,* 2014; Ooi, 2012; Porqueddu *et al.*, 2016). In the Mediterranean region, more frequent and intense droughts are expected, along with an increasing inter-annual variability (Giorgi and Lionello, 2008; Hoerling *et al.*, 2012; Kovats *et al.*, 2014). Widespread devastating droughts and heat waves like those in the summer of 2003 (Ciais *et al.*, 2005) are likely to occur more frequently.

More drought will decrease grassland yields and shorten the grazing season unless the grassland is irrigated (Giannakopoulos *et al.*, 2009; Del Prado *et al.,* 2014), and pasture establishment failures and long-term degradation due to drought are expected to occur (Briske *et al.*, 2003). However, the grassland-based farming system history in Mediterranean Europe shows that these agro-ecosystems have been highly resilient in the face of many changes in climate through several thousand years (Hopkins, 2012). As CO_2 is often a rate-limiting factor in photosynthetic CO_2 assimilation, elevated atmospheric CO_2 concentration has the potential to increase biomass production of C3 plants (Ainsworth and Long, 2005; Sousanna and Lüscher, 2007). However, when N is limiting the positive effect of high atmospheric CO_2 in terms of yield is constrained due to photosynthetic acclimation. Legumes can counteract this limitation by providing N to non-fixing plants in the plant community (Sousanna and Hartwig, 1996; Picon-Cochard *et al.,* 2004). In a Mediterranean climate, water availability will often be more limiting for photosynthesis than CO_2 concentration, but elevated CO_2 can partly compensate for the reduced CO_2 influx through stomata under moderate drought (Ainsworth and Long, 2005; Soussana *et al.,* 2010). Consequently, elevated CO_2 enhances biomass production and improves water relations under drought (Clark *et al.*, 1999), alleviates the drought response in a range of grassland species (Morgan *et al.*, 2004; Aranjuelo *et al.*, 2005; Naudts *et al.*, 2013) and mitigates the effect of extreme climate events on plant growth and production (Zinta *et al.*, 2014). Recent results suggest that in the predicted near future climate, elevated CO_2 could mitigate the effects of extreme droughts and heat waves on ecosystem net carbon uptake (Roy *et al.*, 2016). Elevated CO_2, temperature and drought are commonly observed to increase the competitive ability of legumes in legume-grass mixtures as well as the biological N-fixation as such (Schenk *et al.*, 1995; Sousanna and Lüsher, 2007). This may compensate for the slight decrease in protein content of grasses at elevated CO_2 (Sousanna and Lüscher, 2007; Dumont *et al.*, 2015). However, generalizations about the responses of grassland plant functional types (e.g. legumes and grasses) to elevated CO_2 should be treated with caution. Indirect effects of climate change on species composition and phenology may play a larger role than the direct effect on plants (Dumont *et al.*, 2015). In a glasshouse study, Bolger *et al.*, (1997) found that the variation of growth response to elevated CO_2 by grassland species of the same functional type was much greater than the mean variation between functional types. As a result, grassland compositional change under elevated CO_2 can be expected to depend critically on the specific species' responses (which we do not know much about for most species) in the sward. In a

French low-intensity grassland, simulated warming and summer drought projected for 2080 outweighed the effects of corresponding elevated CO_2, so that aboveground net primary production decreased when temperature, rainfall, and CO_2 were simultaneously manipulated (Cantarel *et al.*, 2013). It was warming rather than elevated CO_2 that increased the observed contribution of legumes to net primary production at the expense of grasses. In any case, with climate change there appears to be an opportunity to better utilize various legume species (Cosentino *et al.*, 2014). Studies on climate change effects on forage quality in Mediterranean grasslands are scarce. However, in a meta-analysis, Dumont *et al.*, (2015) found that unlike other sites, a decrease in herbage N concentration in response to elevated CO_2 was not seen at Mediterranean sites. They suggested that this could be due to changes in species composition (higher legume proportion) and water limitation on growth leading to higher concentration of N.

To summarize, the primary climate challenge in the Mediterranean region of Europe in the future is more severe and frequent droughts. In addition to short-term effects of dry periods on productivity, there will be long-term effects due to a gradual depletion of soil water reserves and changes in species composition in permanent grasslands. There is a high level of uncertainty regarding interactions between climate factors as well as long-term effects.

Climate challenges and opportunities for grasslands in the Nordic region

In the Nordic region, both the observed and predicted warming is more rapid than the global average warming. For instance, in Finland annual average temperatures have increased with more than 2 °C during 1847-2013, almost twice the global average increase. The observed and predicted temperature increase is highest during late autumn, winter and spring (Stocker *et al.*, 2013; Mikkonen *et al.*, 2015), and more frequent warm spells during winter are expected (Johansson *et al.,* 2011). The thermal growing season has increased with about 1-2 weeks during the last 30 years in Norway, and has been predicted to become 1-3 months longer in Norway and Finland by the end of the century as compared to the period 1971-2000 (Ruosteenoja *et al.*, 2011; Hanssen-Bauer *et al.*, 2015). Annual precipitation in the Nordic region is predicted to increase by 5-13% by the end of century relative to 1961-1990, with more frequent episodes of extreme precipitation, especially during winter (Lehtonen *et al.*, 2013; Hanssen-Bauer *et al.*, 2015). Although drought stress is not expected to gain the same importance as in other parts of Europe, drought events are thought to occur more frequently in the future. Decrease in long-term mean snowpack has been predicted toward the end of the century, although individual snow-rich winters will still occur (Räisänen and Eklund, 2011). Thus, there will be shorter duration of snow cover and eventually snow-free winters in some regions now characterized by stable snow cover. On the other hand, increased precipitation in areas where temperatures remain below freezing can give longer-lasting snow cover in some areas (Johansson *et al.*, 2011). Less snow cover can increase the occurrence, depth and duration of soil frost due to less insulation (Bjerke *et al.*, 2015; Kellomäki *et al.,* 2010). The combination of increased precipitation in the autumn and winter, milder and unstable temperatures, and frozen soils, may lead to ice cover or waterlogging.

The short growing season generally limits agricultural production in the Nordic region (Peltonen-Sainio *et al.*, 2009). Thus, longer growing season is favourable, especially in spring when water supply and solar radiation are optimal for growth. In a modelling study, Höglind *et al.*, (2013) found 11% increase in annual dry matter yields of timothy (*Phleum pratense* L.) in northern Europe in 2040-2065 as compared to 1960-1990, with the largest increases in the western regions. This study assumed that timing and number of harvests were adjusted, but did not take the effect of elevated CO_2 on growth into account. In a similar study of timothy and lucerne (*Medicago sativa* L.) mixtures in eastern Canada, which did take the effect of elevated CO_2 into account, estimated 5-35% increase in dry matter yield in 2020-2079 relative to 1971-2000 (Thivierge *et al.*, 2016). In managed grasslands the increasing nutrient requirement can be met through fertilization or inclusion of legumes in mixtures, but in semi-natural grasslands, the positive effect on yield can be offset by declines in soil nutrients, particularly N (Leakey *et al.*, 2009). Most of the dairy production in the Nordic region is based on intensive production systems, and temporary leys constitute a relatively large proportion of grasslands compared to other regions of Europe. Natural grasslands play a significant role in lamb meat production in some Nordic regions (Helgadottir *et al.*, 2014). An extended growing season could mean that less stored forage would be required over the winter, which may lead to a small shift in the direction of less harvested relative to grazed grasslands. However, there are large uncertainties in the predictions, and it may not be possible to utilize all of the extended thermal growing season. Although temperatures increase, the unique photoperiod in Northern latitudes remain unchanged, with rapidly decreasing photoperiod and irradiance from the autumn equinox onwards. Shorter days and low inclination of incoming solar radiation in autumn can limit the amount of photosynthetically active radiation to the point that it becomes a restricting factor for growth, particularly at higher latitudes (Uleberg *et al.*, 2014; Virkajärvi *et al.*, 2015). Moreover, in the current Nordic climate, excessive precipitation frequently causes problems with farm operations in spring and autumn (Peltonen-Sainio *et al.*, 2009, Olesen *et al.*, 2011). Increased precipitation and waterlogged soils could make establishment of new leys, application of fertilizer, and harvesting challenging in some years. Higher temperatures lead to faster decline in the digestibility of forages (Thorvaldson *et al.*, 2007; Bertrand *et al.*, 2008; Bloor *et al.*, 2010), although this effect can be compensated to some extent by altering harvesting regimes (Thivierge *et al.*, 2016), and choice of species and varieties.

Harsh winters are the strongest limiting factor determining which perennial species can be cultivated in the Nordic region. A longer growing season and milder winters may increase the prospective for using species and cultivars with higher yield potential and feeding value, than those used currently. Currently only very winter hardy species, such as timothy, meadow fescue (*Festuca pratensis* Huds.) and red clover (*Trifolium pratense* L.), are widely used (Bélanger *et al.*, 2013, Helgadottir *et al.*, 2014). Especially strong expectations have been posed towards perennial ryegrass (*Lolium perenne* L.), festulolium (×*Festulolium* Aschers. et Graebn.), tall fescue (*F. arundinacea* Schreb.), white clover (*T. repens* L.) and lucerne. Some of these species are currently used e.g. in Denmark, southwestern Norway and southern Sweden, and their use further north would add to feeding value, regrowth ability and productivity of swards. There are, however, many uncertainties regarding winter stresses in a future climate (Rapacz *et al.*, 2014). The type and severity of winter stresses depends not only on the minimum temperature during winter, but also largely on the presence or absence of a snow cover, and on factors that control the cold acclimation status of the plants. Using a modelling approach, Thorsen and Höglind (2010) concluded that for most locations in Norway the risk of winter damage is likely to be reduced in the future. At some locations, however, the risk increased due to a dramatic decrease in snow cover. In winter, when solar irradiation is insufficient for photosynthesis in northern latitudes, the carbon-economy of the plants becomes increasingly important when temperatures exceed 5 °C and respiration increases. On the other hand, if temperatures remain cool, shorter winters could leave more C and N reserves in spring, increasing DM production (Jing *et al.*, 2013; Piva *et al.*, 2013). Cold acclimation of the plants will be delayed to a time when less light is available. This can have impacts on growth cessation and cold acclimation of plants (Østrem *et al.,* 2014; Dalmannsdottir *et al.*, 2015), rendering them more vulnerable to winter stresses. In addition, waterlogged soils in combination with higher autumn temperatures have negative effects on cold acclimation of timothy (Jørgensen *et al.*, 2016). Unstable winter temperatures and early springs can cause plants to de-acclimate, when there is still a risk of freezing (Jørgensen *et al.*, 2010; Rapacz *et al.*, 2014). The distribution of many weed, pests and pathogens that are active in the growing season are limited to the north by harsh winters. With global warming many of these species are expected to spread into or within the Nordic region (e.g. Kaczmarek-Derda *et al.,* 2014). Cultivation of whole crop maize (*Zea mays* L.) has increased during last decades and the expected climate changes will further promote this development, at least in southern parts of the Nordic region (Elsgaard *et al.*, 2012). This could be compatible with an increased use of legumes and total mixed ratio feeding strategies, where high protein forages can be complemented with low protein but high energy components, and thereby influence what type of grasslands are cultivated.

To summarize, altered and unstable precipitation patterns will cause climate challenges in the Nordic region, while there is a high level of uncertainty regarding the effect of climate change on winter survival. Longer thermal growing seasons, elevated CO_2, and the possibility to use other forage species may offer opportunities to increase productivity. The complex interactions between temperature, photoperiod and precipitation throughout the annual growth cycle and winter survival are likely to play a role in determining both challenges and opportunities.

Role of management in adaptation of grasslands in Mediterranean Europe

Using species and variety mixtures where components with desired traits and complementary niches are mixed, is considered a key strategy to maintain production in unpredictable and unstable environments under Mediterranean conditions (Maltoni *et al.,* 2007; Volaire *et al.,* 2014). For example, mixtures of summer-dormant and summer-active perennial species may provide stable pastures exploiting available soil moisture throughout the year (Norton *et al.,* 2016). Grass-legume mixtures including both annuals and perennials proved to achieve higher yields and utilize a longer period for growth, than pure stands (Porqueddu and Maltoni, 2007; Maltoni *et al.,* 2007). Mixed swards are also expected to deal better with climatic variability and to show higher resilience (Lüscher *et al.*, 2014). As described above, the use of legumes in sown mixtures can be expected to have a positive effect on productivity, forage quality and grassland resilience under the projected increase of atmospheric CO_2 and temperatures in the Mediterranean Europe. However, obtaining and maintaining stable species mixtures is not an easy task and further studies are still needed for Mediterranean environments (Porqueddu *et al.,* 2016). Moreover, legumes require relatively high amounts of K and P, and nodulation and N fixation may become limited by low nutrient supply and high temperature (Irigoyen *et al.,* 2014).

Under Mediterranean conditions, grazing by livestock is recognized as the main driver influencing vegetation dynamics and distribution, and grassland productivity (Sternberg *et al.,* 2015). Under climate change conditions, the choice of the most suitable grazing regime will therefore have large effects on soil seed bank dynamics and, in turn, on grassland production. Sternberg *et al.* (2003) found that heavy grazing pressure was unfavourable to the seed bank of annuals if prolonged during seed set, and this negative impact might worsen under future climate scenarios characterized by more intense drought and increased temperatures. There are few studies available on the potential of grazing management strategies in the adaptation of Mediterranean grasslands to climate change. However, key strategies might be to reduce stocking rates, alter rotational grazing systems, and modify timing and period of grazing with the aim of effectively matching animal nutritional requirements with reduced and seasonally shifted pasture production (Köchy *et al.,* 2008; Carmona *et al.,* 2012; Joyce *et al.,* 2013; Pahl *et al.,* 2016). It is advisable to adopt flexible grazing management schemes that take species and functional diversities and forage productivity into account simultaneously, and adapt the level of grazing pressure to water availability (Carmona *et al.,* 2012).

Adaptation to foreseen increased drought may also involve transportation of feed between farms and regions. Complementary forage resources could be produced in the more fertile areas with sufficient water supply (e.g. areas that can be irrigated) and then moved to the more marginal and drought-sensitive livestock areas. In this context, the choice of adapted species and varieties is of primary importance. A wide range of forage species is currently available in the market showing adaptability to Mediterranean climate conditions, such as annual forage species (e.g. *Avena sativa* L., *L. multiflorum* Lam., *T. incarnatum* L., *T. alexandrinum* L., *Vicia sativa* L.), while only very few perennials show good adaptation capacity (Annicchiarico *et al.,* 2013). Another way of creating synergies between regions is transhumance from the plains to the hills and mountains in late spring and vice versa in autumn (Cosentino *et al.,* 2013), but in many areas of Mediterranean Europe has almost disappeared (Azcarate *et al.,* 2013). Promoting this grazing management strategy would be a big challenge but is likely to be effective in a climate change context. The exploitation of alternative forage resources in wooded grasslands could also be a strategy to cope with the foreseen reduced pasture production (Moreno and Pulido, 2009; Del Prado *et al.,* 2014). Such alternative forages may include tree leaves and shrubs, which can alleviate feed shortages, or even fill feed gaps in the winter and especially in the summer in small-scale livestock farms in dry to semi-arid climates (Papanastasis *et al.,* 2008).

Role of management in adaptation of grasslands in the Nordic region

The expected increase in productivity and rate of phenological development in grasslands requires adaptation of management practices in the direction of more frequent harvesting/grazing and increased fertilization levels or more use of legumes. Höglind *et al.* (2013) predicted that the earlier spring and higher temperatures will allow for one more cut per growing season during the future period 2040-2065 compared to 1961-1990. Farmers will, however, take the additional economic costs with an extra harvest into account, and possibly increase grazing instead. With increased precipitation levels, care will need to be taken to minimize soil compaction during farm operations, particularly on some soil types. Old drainage systems may need to be renovated in order to maintain productivity.

Unstable conditions with large variation in weather within and between seasons increase uncertainty in forage production. Diversity among responses to critical weather factors improves resilience at both sward and farm level (Mäkinen *et al.*, 2015). Seed mixtures could exploit temporal and spatial variation in environmental conditions, and be more stable and robust compared to monocultures or simple mixtures. Species and variety mixtures also tend to be more stable in forage quality (Sleugh *et al.,* 2000; Sanderson, 2010), and therefore allows for some flexibility in harvest times, which is desirable in rainy summers. Including N-fixing legumes in such mixtures has several advantages, and can facilitate a better exploitation of elevated atmospheric CO_2 concentration (described above). The conservation of legume forage can be challenging, particularly in a wet Nordic autumn climate, and the proportion of legumes in the mixtures may have to be controlled carefully. Moreover, N fixation may be limited by low temperatures in parts of the growing season (Nesheim and Boller, 1991). Species like perennial ryegrass and festulolium are of increased interest for the future climate, particularly in the Western part of the region, because of their high production capacity throughout the growing season as well as high nutritive value. Timothy-dominated mixtures are still likely to be the main choice in most Nordic regions, and, winters may still be too harsh for the use of perennial ryegrass-dominated mixtures to expand north- and eastwards (Höglind *et al.*, 2013). Including deep-rooted drought tolerant species like tall fescue and fescue type festulolium, red clover and lucerne in the mixtures can be a strategy to prevent yield reduction during drought periods. This is particularly relevant for the drier inland and eastern parts of the region. Tall fescue has a better nitrogen utilization and water use efficiency, and a yield potential which is up to 50% higher than perennial ryegrass in dry periods, most probably owing to its deeper rooting system (Cougnon, 2013). In addition, it can tolerate poorly drained soils (Barnes *et al.,* 2003). However, the digestibility and animal preference is lower than in perennial ryegrass (Cougnon *et al.*, 2014).

Exploitation of plant traits in adaptation of grasslands in Mediterranean Europe through breeding

Drought escape (i.e. plants survive the dry summer as seeds; Long *et al.,* 2014) and hardseededness (which allows a more persistent seed bank; Taylor, 2005) are the main adaptive strategies of annual species which dominate in Mediterranean grasslands. With the predicted changes in rainfall distribution, with an overall reduced growth period (variable onset of autumn rainfall and shorter spring), it has been suggested to promote the seeding of annual species characterized by the following traits (Porqueddu *et al.,* 2016 and reference therein): (2) earlier seed maturation; (2) higher proportion of hard seeds to prevent seedling losses from more prevalent false breaks; (3) an indeterminate or less determinate flowering habit, which gives flexibility to exploit better growing seasons.

Regarding perennials, species or populations with greater drought tolerance will be needed (Olesen *et al.*, 2007). However, breeding efforts in 'cool season' forage plants have taken place mainly in temperate areas and very few cultivars adapted to severe drought are currently available in Europe (Lelièvre and Volaire, 2009). Persistence during severe drought is governed by mechanisms different from those conferring resistance to moderate droughts (Milbau *et al.*, 2005; Volaire *et al.*, 2009). Plant responses resulting in resistance under moderate drought through the maintenance of aerial growth have to avoid and/or tolerate leaf dehydration. At moderate drought, the maintenance of biomass production can be achieved primarily by maximising soil water capture while maintaining stomatal gas exchange and transpiration (Blum, 2009). A deep root system with a high density of roots at depth (Carrow, 1996; Wasson *et al.*, 2012; White and Snow, 2012) and maintenance of leaf area, leaf relative water content, leaf cell turgor and photosynthetic capacity (Morgan, 1988; Serraj and Sinclair, 2002) are traits that are associated with high yield in water-limited environments. Plant responses resulting in survival under severe drought, however, are mainly associated with dehydration avoidance and tolerance occurring in young tissues including basal meristematic tissues. When conditions improve, the surviving meristems can generate new leaves if the adult leaves are dead (Van Peer *et al.,* 2004; Zwicke *et al.*, 2015). In some species and genotypes from very dry areas, survival of basal meristematic tissues is achieved through summer dormancy (Volaire and Norton, 2006). In these plants photoperiod and temperature induces (even under irrigation) cessation or reduction of shoot growth, various degrees of foliage senescence and a dehydration tolerance of meristems. The reduction in leaf tissue reduces total plant water loss (Gepstein, 2004; Munne Bosch and Alegre, 2004). A minimum water supply to the meristematic tissues is maintained (Karcher *et al.*, 2008; McWilliam and Kramer, 1968; Volaire and Lelievre, 2001), and high concentrations of fructans and dehydrins contribute to osmoregulation and membrane stabilisation of these tissues (Hincha *et al.*, 2000; 2002). High carbohydrate reserves are associated with superior plant resilience and recovery after severe drought, with a strong interaction with defoliation intensity (Boschma *et al.*, 2003). Thus, to interpret low leaf water potential or high foliage senescence as responses associated with drought sensitivity and poor adaptation may be correct if maintained production under drought is the target, but highly misleading if drought survival during severe drought is the focus. Making the distinction between the responses of mature and young meristematic tissues is crucial when analysing the strategies of perennial grasses to contrasting drought intensities. It may be possible to combine drought resilience of perennial forage species with high biomass productivity in rainy seasons, as recently shown by crossing summer dormant with summer active and highly productive genotypes of cocksfoot (*Dactylis glomerata* L.) (Kallida *et al.,* 2016).

Exploitation of plant traits in adaptation of grasslands in the Nordic region through breeding

In order to utilize a longer growing season, higher temperature and higher CO_2 levels in the future, we need species and varieties that have a strong regrowth capacity and tolerate more frequent harvesting or grazing. However, resistance to winter stresses are still likely to be of high importance (Rapacz *et al.*, 2014). Plants encounter many stresses during winter: freezing, anoxia due to ice encasement or water-saturated soils, soil movements due to freeze-thaw cycles, winter pathogens, starvation and dehydration due to frozen soils. Specific resistance mechanisms to these stresses exist, but they are also largely interconnected through genetics and physiology. Central to winter survival is proper cold acclimation in autumn, and maintenance of a cold acclimated state for sufficiently long in spring. During cold acclimation, leaf elongation ceases, a number of stress responses are elicited, and organic reserves are accumulated. Both cold acclimation and deacclimation are mainly controlled by temperature. However, light is also important in at least three different ways: (1) high irradiance combined with low temperature increases the photosystem II excitation pressure, eliciting signalling pathways leading to cold acclimation; (2) photoperiod is a developmental signal influencing growth, cold acclimation and deacclimation; and (3) light is the energy source for the accumulation of organic reserves. Due to these reasons, the shift of cold acclimation and deacclimation into shorter photoperiods may affect the ability of plants to cope with winter stresses. A more detailed and quantitative understanding of interactions between temperature and light on winter survival in different species is needed. Breeding activities may need to focus on adjusting the timing of growth cessation and cold acclimation in autumn, and the opposite process in spring, to new temperature and photoperiod combinations. For example, at high latitudes, perennial ryegrass and festulolium tend to cease growth in autumn too late for sufficient cold acclimation (Østrem *et al.*, 2014). It will, however, be necessary to manage the trade-off between optimal timing with respect to winter survival, and utilization of the longer growing season to increase production. For timothy, the priority would likely be to improve regrowth capacity and spring growth to exploit favourable conditions in spring/early summer, while for tall fescue, work is ongoing to combine the high yield and drought tolerance with an acceptable digestibility and animal preference (Humphreys *et al.*, 2012, Cougnon *et al.*, 2015; Fariaszewska *et al.*, 2016). A broadening of the gene pools of all species is necessary, and future breeding, irrespective of crop species, demands efficient ways to incorporate wild adapted genetic resources and exotic material into the current breeding base. This is a long-term task which requires public support and active knowledge transfer from public sector scientists into real-life crop improvement (Helgadóttir *et al.*, 2014).

Conclusions

In the face of unstable and uncertain climatic conditions, a high diversity of cultivated forage species, high intraspecific genetic diversity, and the use of species and variety mixtures will positively affect productivity and resilience of grasslands. Legumes are likely to contribute to resilience and productivity due to their ability to maintain persistent seed banks, utilize elevated CO_2 and prevent N limitation of companion grass growth. In both the Mediterranean and Nordic regions, climate change will lead to changes in the annual growth cycle of grassland species, prompting a range of adaptations of management practices. Breeding efforts should be directed both towards improving plant strategies to cope with abiotic stresses, and towards adaptation to new seasonal patterns and management, while minimizing possible trade-offs with productivity. A more detailed understanding of interactions between ambient CO_2 concentration, precipitation, temperature, photoperiod, irradiance and management on drought resistance, winter survival, species composition, productivity and forage quality would aid this process.

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Priorities for modelling European grasslands under climate change

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Abstract

Climate change presents a series of interacting challenges for grassland-based livestock systems in Europe. Modelling is an invaluable tool for understanding how complex systems such as grasslands may respond to future change, and how these responses might be mediated by adaptation and mitigation strategies. In order to understand the challenges to, and research priorities for, European grassland modelling under climate change, a 'horizon scanning' approach was used to gain the views of grassland modellers and researchers from across Europe. Participants assessed the current state of grassland modelling, identifying the challenges faced by modellers, and the steps needed to overcome them. Qualitative analysis revealed a common structure underlying the research priorities, which could potentially support the focussing and structuring of future funding frameworks and projects. For five challenges, the importance of new experimental research was identified as a priority for developing modelling capacity.

Keywords: climate change, collaborative research, grassland models, research priorities

Introduction

Grassland-based livestock systems represent a vital component of European agriculture, producing a range of products and providing a number of important societal benefits. In the context of climate change, population growth and changing dietary preferences, livestock production must become more efficient while maintaining sustainability (Thornton, 2010). To meet these challenges, diverse, complex grassland production systems need to be better understood, and measures to adapt to global change need to be tested. Modelling can play an important role in this respect, in that models can reveal interactions, depict hidden processes and allow policies and management options to be explored without risk to real systems (Van Paassen *et al.*, 2007). However, the capacity to meet the challenges posed by global change can be limited if researchers in different nations and institutes, and those involved in modelling different aspects of grassland systems, do not learn from each other or effectively communicate their requirements to the wider scientific and policy community (Kipling *et al.*, 2014). In 2012 the Agriculture, Food and Climate Change Joint Programming Initiative (FACCE JPI) of the European Union set up the Modelling European Agriculture with Climate Change for Food Security (MACSUR) project (http://macsur.eu) to build capacity in agricultural modelling (Kipling *et al.*, 2014). In the frame of MACSUR, this study presents and analyses the priorities for grassland modelling in Europe under climate change.

Materials and methods

In June 2015 a workshop was held at Wageningen UR (the Netherlands) and a questionnaire distributed to identify challenges and research priorities for modelling European grasslands in the context of climate change. A 'horizon scanning' approach, using the 'Futures workshop' method (Jungk and Müllert, 1987; Valqui Vidal, 2005) was used to gain the views of grassland modellers and researchers from 16 institutes in 10 countries. The process identified three types of challenge: (1) ten challenges relating to individual climate change impacts (Figure 1); (2) two cross-cutting topical challenges (modelling different regions

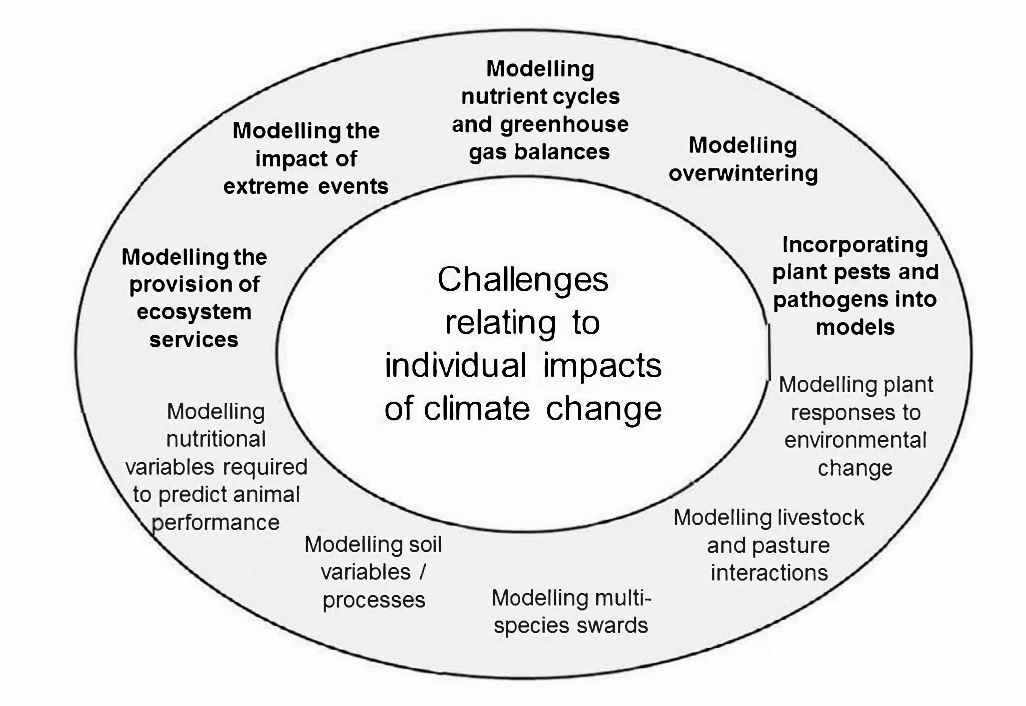

Figure 1. Challenges for grassland modelling relating to individual climate change impacts. Bold type indicates the five challenges in which experts prioritised the need for new experimental work.

and production systems, and adaptation to climate change); and (3) three cross-cutting methodological challenges ('fit for purpose' modelling, linking scales of modelling and data, providing data for modelling). The current paper focussed on group (1), and compared the research priorities identified in these challenges. Comparisons were made using a qualitative approach (Ritchie *et al.*, 2014) involving coding the text of the synthesized outputs from original workshop and questionnaire responses. The analysis aimed to identify commonalities in the underlying structure of the identified research priorities.

Results and discussion

Analysis of the 10 priorities for modelling relating to individual climate change impacts revealed a common underlying structure (Figure 2). The three identified methodological challenges map onto the categories revealed (for example, 'providing data for models' relates to 'inventory', 'external progression', and 'using external outputs'). Exploration of the categories described can help inform the structuring of research frameworks for agricultural modelling. Future analysis will examine research priorities for other fields of agricultural modelling (e.g. livestock disease), to further develop and test the suggested categorisation, and to compare the categories identified with existing theories of knowledge development.

The categories described (Figure 2) provide a framework for progress in each of the 10 challenge topics relating to individual climate change impacts. For five challenges, the need for more experimental research (categories 'external progression' and 'acting together') was identified as a priority (Figure 1). This does not imply that for other challenges new experimental data would not be valuable, but for these, other advances (developing models using existing under-utilised data, or assessing data requirements) were more emphasized.

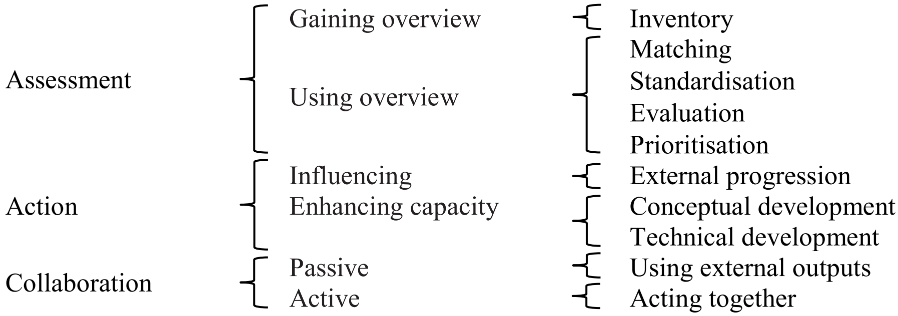

Figure 2. Nested levels of categories relating to research priorities for grassland modelling.

Conclusions

This study identified research priorities for European grassland modelling in the context of climate change, and described a general framework for research progress. Key components of addressing the research priorities identified were assessment of and action to enhance experimental and modelling capacity, and collaboration across disciplines.

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Intercomparison of models for simulating timothy yield in Northern countries

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Abstract

Simulation models are widely used to assess the impact of climate change on crop production and adaptation options, but few model comparisons have been done to assess uncertainties in the simulation results of forage grass models. The aim of this study was to compare the performance of three models (BASGRA, CATIMO, and STICS) to simulate the dry matter yield of the first and second cut of timothy (*Phleum pratense* L.) using observed field data from a wide range of climatic conditions, cultivars, soil types and crop management practices that are associated with timothy production in its main production regions in Canada and Northern Europe. The performance of the models was assessed with both cultivar-specific and non-cultivar-specific (generic) calibrations. The results showed the strengths and weaknesses of different modelling approaches and the magnitude of uncertainty related to simulated timothy grass yield. Model results were sensitive to calibrations applied.

Keywords: model comparison, *Phleum pratense* L., simulation, timothy, yield

Introduction

Model-based decision support tools are increasingly used to support crop management planning in agriculture and to anticipate the impact of climate change on crop production. Process-based models are thought to be powerful tools for those purposes as they aim to describe the mechanisms of complex interactions between crops and their environment. Forage grass production systems have also been studied with such tools (Höglind *et al.,* 2013; Jing *et al.,* 2014).

During the last few years, several model comparison studies have been published for cereal crops. The Agricultural Model Intercomparison and Improvement Project (AgMIP) (www.agmip.org) and the MACSUR knowledge hub (www.macsur.eu) have used crop model comparisons to identify crop model weaknesses that hamper climate impact assessments. Model comparisons have been published for forage grasses too (e.g. Hurtado-Uria *et al.,* 2013), but these have often been restricted to small regions with homogeneous climate conditions and none have been done for timothy, one of the most important forage grass species in the cold temperate climate zone of the northern hemisphere, including Canada, and the Nordic countries in Europe.

Our objective was to compare the performance of three growth models (BASGRA, CATIMO, and STICS) to simulate the dry matter (DM) yield of the first and second cut of timothy using observed field data from a wide range of climatic conditions, cultivars, soil types and crop management practices that represented the main production regions of timothy in Canada and Northern Europe. The performance of the models was assessed with both cultivar-specific and non-cultivar-specific (generic) calibrations.

Materials and methods

The three models included in the comparison were CATIMO (Bonesmo and Belanger 2002), BASGRA (Höglind *et al.,* 2001, Van Oijen *et al.,* 2005), and STICS (Jégo *et al.,* 2013). For the comparison of the models, we chose data from field trials at seven sites across Northern Europe and Canada (Table 1).

Table 1. Study sites and cultivars.

Study site	Cultivar	Years
Fredericton, Canada	Champ	1991-1993
Lacombe, Canada	Climax	2004-2005
Maaninka, Finland	Tammisto II	2006-2007
Quebec, Canada	Champ	1999-2001
Rovaniemi, Finland	Iki	1999-2001
Særheim, Norway	Grindstad	2000-2002
Umeå, Sweden	Jonatan	1995-1996

Data from two to three growing seasons per site were available, and they covered altogether 33 different treatments with, for example, varying nitrogen application rates and cutting dates.

Model calibrations were first carried out for individual cultivars, and then one generic calibration was done using the data from all study sites. Altogether, 24 treatments were used for calibration and 9 randomly selected treatments per management type and site for sites with more than 2 treatments for model performance evaluation. Model input data for each site consisted of daily weather (i.e. minimum and maximum temperatures, precipitation, relative humidity, wind speed, and global radiation), crop management (cutting dates and fertilisation dates and amounts) and information on soil including texture, organic matter, bulk density and hydraulic properties. In addition, field observations (i.e. total aboveground dry matter, dry matter of leaves and stems, crop height, leaf area index, specific leaf area and tiller density) were provided.

The performances of the models, i.e. their capacity to predict dry-matter yield of the first and second cuts, were evaluated for both cultivar-specific and generic calibrations.

Results and discussion

The three models generally simulated DM yields at different sites fairly well (Figure 1). The simulation accuracy for calibration treatments was slightly better than that for the treatments used for evaluating model performance: RMSE was from 10 to 35% smaller for calibration treatments depending on model. With cultivar-specific calibration, for the spring growth and the resulting first cut, STICS simulated the DM accumulation better than BASGRA and CATIMO (RMSEs of evaluation treatments were 80, 150 and 140 g m^{-2}, respectively). However, BASGRA performed better for the regrowth and the resulting second cut (RMSEs 60, 130 and 210 g m^{-2} for BASGRA, STICS and CATIMO, respective). The cultivar-specific calibration resulted in better simulation accuracy than the generic calibration: RMSE was from 16 to 24% smaller for cultivar-specific calibration depending on model.

Improved understanding of the performances of the models and related uncertainties is essential for model use in climate change impact assessments. All models managed to estimate the yields satisfactorily and none of them worked clearly better than others. The evaluation of model performance for forage nutritive quality remains an important aspect to be included in model comparisons.

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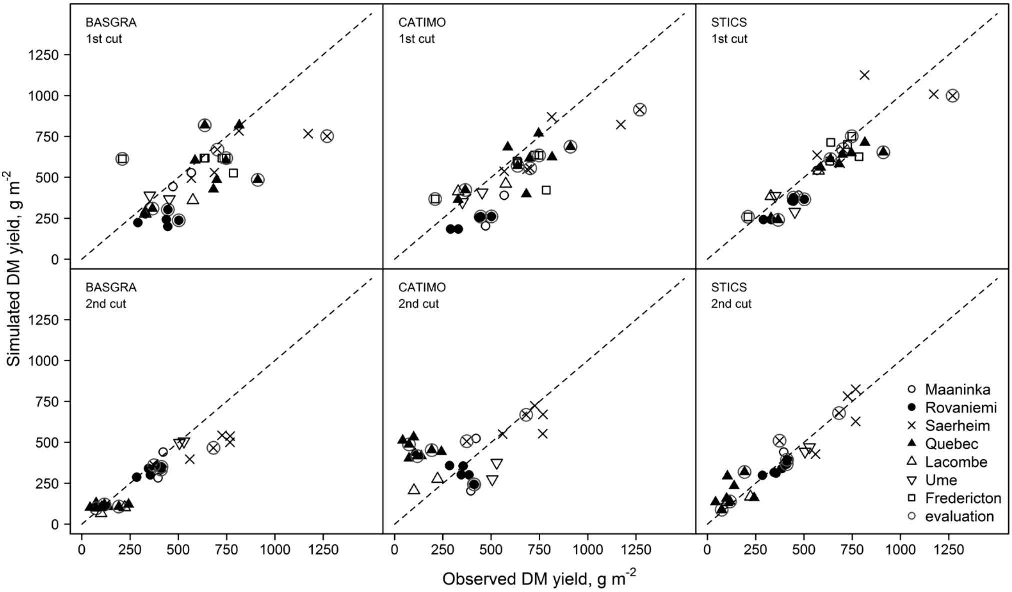

Figure 1. Simulated and observed yield estimates of 1st and 2nd cuts (g m^{-2}, dry matter) for 33 treatments for the three timothy models using cultivar-specific calibration. Different study sites are depicted with different symbols and treatments used for evaluation are circled in grey. The dashed 1:1 line represents perfect agreement.

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Nitrogen is a key indicator for sustainable use of European grasslands

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Abstract

High input grassland systems are characterized by (high) surpluses of nutrients like nitrogen, brought in from synthetic fertilizers, concentrates, and sometimes external produced animal feedstuffs. Nitrogen is in most cases the main responsible actor for high emissions of nutrients to air, soil and water. Nitrogen balances and NUE (nitrogen use efficiency) are therefore useful as indicators. Low input grassland systems sometimes suffer from overgrazing which results in decreasing grass yields and lower soil cover with grasses. Nitrogen is not the main responsible actor for overgrazing, but nitrogen balances and NUE are useful as indicators to improve sustainable use of low input grassland systems. The paper shows that in general the sustainability of high input grassland systems will be improved by lowering the nitrogen balance surplus and by increasing NUE. It is shown that system boundaries of farming systems have to be taken into account before reaching final conclusions.

Keywords: animal feed, animal manure, animal production, nitrogen surplus, nitrogen use

Introduction

The total agricultural area in the EU-27 member states comprises 190 million hectares, divided in 121 million hectares arable land and 69 million hectares permanent grassland. Grassland based agricultural production is responsible for large environmental emissions causing eutrophication and climate change. High input grassland systems are characterized by external inputs of synthetic fertilizers, concentrates, and sometimes external produced animal feedstuffs. These systems often have high surpluses of nutrients like nitrogen, resulting in considerable emissions in the form of ammonia, nitrate and nitrous oxide. Low input grassland systems are characterized by low external inputs. These systems sometimes suffer from overgrazing which results in decreasing grass yields and lower soil cover with grasses. Overgrazing leads to more sensitivity to soil erosion (both water and wind erosion). In the long term, overgrazing leads to lower carbon storage in the soil due to reduced plant growth. This paper focuses on measures for sustainable grassland based animal production systems and on the use of nitrogen indicators to monitor the effect and implementation of these measures.

Materials and methods

A nutrient budget sheet keeps track of all inputs and outputs from a farm and allows calculating the nitrogen use efficiency (NUE) and the nutrient surplus. In this paper the so-called farm gate budget sheet for nitrogen is used, with the following system boundaries and definitions:

FI = external farm inputs: synthetic fertilizer, purchased animal feedstuffs, purchased animal manure, biological nitrogen fixation (BNF), nitrogen deposition

FP = farm products: meat, milk, wool, eggs, sold crops, sold animal manure

NUE = FP/FI

N surplus = FI – FP

Table 1. Effect of some important measures on nitrogen use efficiency (NUE) and N surplus, on individual farms.

Measure	NUE_{farm}	N $surplus_{farm}$
1. Feeding dairy cows according to feeding standards	increase	decrease
2. More import of externally produced animal feed	increase	increase
3. Shortening the grazing period of dairy cattle	increase	decrease
4. Better use of animal manure by low ammonia emission storage and application	increase	decrease
5. Replacing synthetic fertilizer by BNF on grassland farms[1]	equal	equal
6. Adapting stocking rates to carrying capacity in rangelands	increase	decrease

[1] BNF = biological nitrogen fixation.

This formulation means that farm grown forages internally used as feed and farm animal manure internally used as organic fertilizer are considered as internal entries which do not enter the budget sheet. Biological fixed nitrogen by forage legumes is considered as an entry on the budget sheet because when the fixed nitrogen becomes part of the farm soil system it cannot be distinguished from synthetic nitrogen fertilizer. So, it contributes in the same way as synthetic nitrogen fertilizer to nitrogen losses. The nutrient budget sheet does not provide information about the distribution of the surplus on the individual components ammonia, nitrate and nitrous oxide (Bleken *et al.*, 2005; Powell *et al.*, 2010; van der Hoek and Bouwman, 1999).

Results and discussion

A number of measures that have been studied in the framework of sustainable animal production systems are presented in Table 1, together with the effect on NUE and N surplus (see for details Bleken *et al.*, 2005; Van der Hoek, 2014).

Measure 1 relates to farms where dairy cows are not fed according to the feeding standards. Feeding below or above the feeding standards means the supplied animal feed is suboptimal used. Adjusting the feed supply to the feeding standards, results in both cases to increasing NUE and decreasing N surplus. Increasing the amount of animal feed by importing more externally produced animal feed as mentioned in measure 2 will also increase NUE because the associated crop production losses are accounted for elsewhere. N surplus increases because the extra feed input causes also losses on the receiving farm in the animal house and during manure application. Shortening the grazing period of dairy cattle in measure 3 means less manure excretion in the meadow and more manure collected in the animal house and this extra amount of animal manure can replace part of the purchased synthetic fertilizer (Van der Hoek, 2010). Measure 4 results in lower ammonia emissions. So more nitrogen is available for the crop and this can replace some of the purchased synthetic fertilizer (Bittman *et al.*, 2014). Measure 5 compares a situation with similar milk and meat yield per hectare for legumes/grassland and grassland with synthetic fertilizer. So there will be no difference in NUE and N surplus (based on Ledgard, 2001; Schils *et al.*, 2000). This measure results in lower synthetic fertilizer consumption. As synthetic fertilizer production is associated with high CO_2 emissions, this is a positive side effect.

Measure 6 improves sustainability of animal production by reducing overgrazing. In rangelands, plant and animal output both will increase when stocking rates are adapted to carrying capacities. This means NUE increases and N surplus decreases (Oliva *et al.*, 2012).

Conclusions

1. Nitrogen budgets are a useful tool, and NUE and N surpluses are useful indicators to monitor the effect and implementation of measures to improve the sustainability of agricultural production systems.
2. The measures collected in Table 1 are all studied on individual farms. Some measures, like 1 and 3 through 6, deal with internal farm management, without impact on other farms. In general these measures lead to increasing NUE and decreasing N surpluses.
3. Some measures taken on individual farms have an impact on other farms. As with measure 2, when individual grassland farms increase their import of external produced crops, they improve their NUE. However when the crop producing farm has to buy additional synthetic fertilizers instead of using the corresponding animal manure, NUE on a national scale will not improve.
4. As a consequence, measure 2 as described above is a plea for mixed farming systems that integrate crops and livestock, with animal manure production and sustainable use of it in a single head of management.
5. Some measures, like 3 and 4, aim at more and better use of animal manure as fertilizer and consequently have savings on synthetic fertilizer. Measure 5, introducing legumes on grassland farms, replaces also synthetic fertilizer. As synthetic fertilizer production is associated with high CO_2 emissions, this is a positive side effect of the use of legumes.

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Fertilizing strategy and spreading technology for cattle slurry – grass yield and ammonia emissions

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Abstract

Swedish nitrogen (N) fertilizer recommendations need to be evaluated to match current grass ley production. The aim of this study was to identify fertilizer strategies for cattle slurry concerning time, applied N amounts and spreading technologies that decrease ammonia losses (NH_3-N) and increase dry matter (DM) yield and N efficiency in ley with three cuts per year. The three-year grass ley trial used a complete randomized block design with four replications on clay soil. In three of the treatments chemical N fertilizer in spring was combined with cattle slurry after the first ley harvest. In one treatment cattle slurry was spread at spring and chemical N fertilizer spread after the first ley harvest. Slurry was applied with bandspreading and shallow injection after first cut and NH_3-N emissions were measured. Shallow injection decreased NH_3 emissions by more than 50% when compared with band spreading, but did not give a significant higher DM yield. On average, N use efficiency was 50% when spreading chemical N at spring and cattle slurry after first cut. This was higher than spreading slurry at spring and chemical N after first cut.

Keywords: grass yield, cattle slurry, nitrogen, ammonia emissions, technology

Introduction

Spreading cattle slurry to grass ley requires consideration of time, applied amounts and technology. These factors influence yield and nitrogen (N) losses, which mainly occur as NH_3-N emissions (Webb *et al.*, 2013). The aim of this study was to identify fertilizer strategies and spreading technologies to decrease NH_3-N emissions and increase dry matter (DM) yield and N efficiency using cattle slurry and chemical N fertilizer for grass ley with three cuts per year.

Materials and methods

A seed mixture of 25% *Lolium perenne* L., 45% *Phleum pratense* L. and 30% *Festuca pratensis* Huds. was used to establish the sward the year before. The three-year field experiment used a randomised complete block design with four replications of each field plot (12×3 m). In three of the treatments chemical N fertilizer in spring was combined with cattle slurry after the first ley harvest (Table 1). In one treatment cattle slurry was spread at spring and chemical N fertilizer spread after the first ley harvest. No N fertilizer was added to the third harvest. The field trial also included an unfertilized treatment and treatments with increasing amount of chemical N fertilizer. Temperature and precipitation was documented during the growing season with a local weather station Vaisala WXT 510. In one of the treatments cattle slurry was spread with shallow injection (c/c distance 0.25 m) in open slots (depth 0.05 m) after first harvest and was compared with band spreading of cattle slurry, c/c distance 0.25 m. Ammonia emissions were measured with a micrometeorological difference method until emissions were negligible, i.e. 60-70 hours after application (Svensson, 1994). Content of N in cattle slurry was analysed and applied amounts were documented and used to calculate N use efficiency and proportion of N lost as NH_3-N. Statistical analysis of DM yields was conducted with mixed linear models, where treatment within years was a fixed factor (SAS Institute Inc., 2008). Yields in each harvest were determined plot-wise and samples of herbage were analysed for DM content and crude protein. Nitrogen yield of the total harvest on an annual basis was calculated for each treatment. Nitrogen yield efficiency was calculated as the ratio of

Table 1. Experimental design of the field trial. Amounts of NH_4-N applied to first and second harvest, kg ha^{-1}.

Treatments[1]	1st harvest	2nd harvest	Total NH_4-N
Control	0	0	0
M/M 80/60	80	60	140
M/M 110/30	110	30	140
M/M 115/95	115	95	210
M/Sb 80/60	80	60	140
M/Sb 110/30	110	30	140
M/Ss 80/60	80	60	140
Sb/M 60/80	60	80	140

[1] M = mineral N in chemical fertilizer; S = cattle slurry; b = band-spreading technology; s = shallow injection technology.

added yield of N when compared to unfertilized treatment and total added amounts of NH_4-N with cattle slurry and chemical fertilizer.

Results and discussion

Shallow injection of cattle slurry resulted in NH_3-N emissions one third as high as band spreading in the second year (Table 2). Third year NH_3-N emissions were halved with shallow injection. Dry matter and NH_4-N contents of cattle slurry at spreading time varied between 5.2-9.3% and 1.6-2.3 g kg^{-1} fresh

Table 2. Application of N with cattle slurry and NH_3-N emission losses.

Year	Treatments	Amounts			Emissions of NH_3-N		
			kg ha^{-1}			percent of	
		m^3 ha^{-1}	total-N	NH_4-N	kg N ha^{-1}	total-N	NH_3-N
2	M/Sb 80/60	24	93	64	31	33	48
	M/Ss 80/60	25	94	65	9	10	14
3	M/Sb 80/60	30	109	49	21	20	44
	M/Ss 80/60	31	112	50	11	10	22

Table 3. Average dry matter yield for three years, Mg ha^{-1}.[1]

Treatments	1st harvest	2nd harvest	3rd harvest	Total harvest
Control	4.8^{a}	2.1^{a}	1.7	8.6^{a}
M/M 80/60	5.6^{b}	3.2^{b}	1.9	10.7^{b}
M/M 110/30	5.7^{b}	3.1^{bc}	1.7	10.5^{b}
M/M 115/95	5.7^{b}	3.1^{bc}	1.9	10.7^{b}
M/Sb 80/60	5.9^{b}	2.9^{cd}	1.9	10.7^{b}
M/Sb 110/30	5.9^{b}	2.8^{d}	1.7	10.4^{b}
M/Ss 80/60	6.0^{b}	2.9^{cd}	1.8	10.7^{b}
Sb/M 60/80	5.6^{b}	2.9^{cd}	1.7	10.1^{b}
LSD (Tukey)	0.51	0.29	0.30	0.79

[1] Average yield with different letters within a column are significant different (P<0.05).

weight and was representative for slurry management systems in dairy production (Steineck *et al.*, 1999). First year measures are not presented due to inappropriate injection.

There were no significant differences between treatments concerning DM yield when combining cattle slurry and chemical N fertilizer (Table 3). However, different growing conditions between years resulting in different impacts on the N use efficiency (Figure 1). First and last growing season was wetter than normal, and first and second growing season was warmer than normal. On average N use efficiency was 50% when application of chemical N at spring was combining with cattle slurry after first harvest. This was higher than spreading cattle slurry at spring and chemical N after first cut.

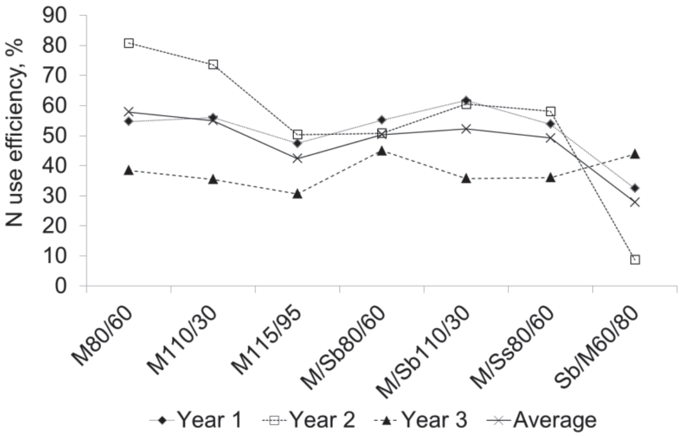

Figure 1. N use efficiency for each treatment and year and average over three years.

Conclusions

Shallow injection of cattle slurry after first harvest more than halved NH_3-N losses compared with band spreading. There was no significant difference in average DM yield between different strategies of combining chemical N fertilizer and cattle slurry. The use of shallow injection gave a similar DM yield as band spreading. About 50% of added N with chemical fertilizer at spring and cattle slurry after first cut was removed with total DM yield.

Acknowledgements

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Greenhouse gas emissions and agronomic feasibility for forage production on inverted peat soil

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Abstract

We studied greenhouse gas (GHG) emissions (CH_4 and N_2O), agronomic performance and soil conditions in a grassland on an inverted peat soil that had earlier been cultivated and tile-drained, and compared it with grassland on conventionally tile-drained peat. A neighbouring undrained peat was used as a reference for GHG emissions. Preliminary results (2-year field data) revealed reduced GHG emissions from the inverted peat relative to the tile-drained peat, mainly caused by lower CH_4 emissions. Our data suggest that peat inversion can improve the agronomic feasibility of forage production in cool-moist areas with abundant organic soils, and can offer a way of agronomic adaptation to a climate with increased precipitation. At the same time it may reduce the GHG footprint of forage production.

Keywords: greenhouse gas mitigation, peat soil, drainage, adaptation

Introduction

Many grasslands in Western Norway are situated on former bogs, posing agronomic and environmental challenges. In some regions, grasslands are established on cultivated tile-drained peat soils situated on top of a self-draining mineral soil covered by a thin layer of impermeable mineral soil. In such regions, peat inversion can be a viable strategy to ameliorate cultivated peatland for forage production, and to protect the peat from further decomposition. The peat soil is covered with mineral soil while maintaining connectivity to the self-draining subsoil by means of tilted mineral soil layers (Figure 1). The objective of the present study was to investigate if peat inversion (IP-soil) could reduce CH_4 and N_2O emissions relative to tile-drained peat (TD-peat), while at the same time improving its agronomic performance. We hypothesised that peat inversion would result in higher grass yields, better soil aeration, more efficient infiltration and drainage, lower ground water tables, and less N_2O and CH_4 emissions, relative to tile-drained peat. To assess the overall effect of peat cultivation on CH_4 and N_2O emissions, an uncultivated peatland was included in the study.

Materials and methods

In 2014, grassland fields were established on an IP-soil that was previously a TD-peat, and a neighbouring TD-peat at Fræna in western Norway (62°96'N, 7°14'E). N fertilizer was applied as 30 tonne cattle slurry (54 kg NH_4-N ha^{-1}) and 150 kg NH_4NO_3-N ha^{-1} in 2014 (establishing year). In 2015, only mineral fertilizer was used (260 kg N ha^{-1}), and split between spring (150 kg N ha^{-1}) and after first harvest (110 kg N ha^{-1}). To be comparable to normal farming practise, the grasslands were compacted with three passes wheel by wheel per year by a tractor (5.2 tonne), as the grasslands were cut with a two-wheel weeper.

We investigated grass yields, soil physical conditions, depth to ground water table, and N_2O and CH_4 emissions throughout two growing seasons. Greenhouse gas (GHG) measurements on uncultivated peat were carried out in 2015, only. All three peat sites were situated in close vicinity to each other on top of a silty sand. N_2O and CH_4 fluxes were measured by closed chamber technique approximately weekly throughout the growing season (starting from summer 2014, when the field was established), with six chambers in each of the two grasslands, and two chambers on the uncultivated peat. During a period

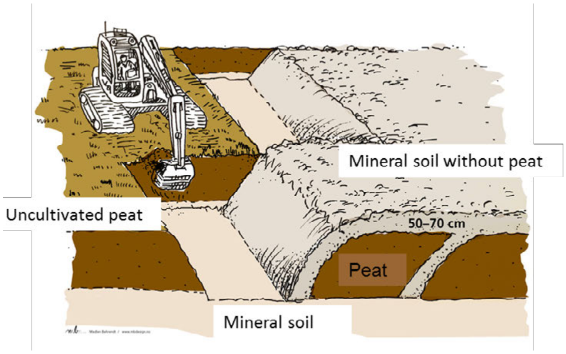

Figure 1. Principle for peat inversion.

of two weeks after fertilizer application, more frequent flux measurements were performed, whereas measurements were less frequent in autumn.

To check for CH_4 accumulation in the inverted peat body, we installed porous suction cups at the same location as the flux chambers in 2015, probing the top of the buried peat layer (95-105 cm below soil surface). Two more cups were installed 20 and 40 cm above this in the silty sand above the peat. Pore size distribution and aeration properties of the topsoil were determined in undisturbed 100 ml cylinder cores taken from 5-10 cm depth. Depth to the ground water table (GWT) was measured at every gas sampling by a tape measure in perforated vertical PVC tubes, permanently installed close to the chambers.

Results and discussion

The total pore volume was largest in the TD-peat, but the air-filled porosity was smaller than in the IP-soil. In the TD-peat the water infiltration after rain was slow, resulting in periods with waterlogged soil. During 2015, GWT in the TD-peat varied between 0 and -117 cm, with an average of -68 cm. In the IP-soil, the GWT in the tilted layers of silty sand were mostly below 130 cm. In contrast to the TD-peat, the top soil of the IP-soil was never waterlogged.

In both fields, the grass yields were high compared with the average in the district, being 14.9 and 11.2 t dry matter per ha on IP-soil and TD-peat, respectively, in 2015. Cumulative N_2O emissions were slightly lower in TD-peat than in IP-soil in the warm and dry summer of 2014 (1.5 versus 2.0 kg N_2O-N ha^{-1} in fertilized plots), whereas in the wet year of 2015, N_2O emissions were greater in TD-peat (4.3 versus 3.6 kg N_2O-N ha^{-1} in fertilized plots; Figure 2). In 2015, N_2O peak emissions in TD-peat were observed shortly after fertilization (max 1,900 μg N_2O-N m^{-2} h^{-1}), whereas IP-soil had smaller peaks that lasted longer (max 330 μg N_2O-N m^{-2} h^{-1}). The reason for the higher fluxes shortly after the fertilization in TD-peat is likely to be the rapid denitrification of added NO_3 in the wet peat, fuelled by easily available carbon from the degrading peat. A similar pattern of N_2O emissions was observed in another poorly drained peat soil in western Norway (Hovlandsdal, 2011). Only a small N_2O emission was observed in unfertilized TD-peat (Figure 2). This may be due to the poor drainage (with periodical waterlogging) and/or low nutrient content of the peat.

CH_4 emissions were much larger in the TD-peat than in the IP-soil. In 2014, CH_4 emissions were high in late autumn, whereas in the wet year of 2015, high emissions were observed throughout the whole summer. There were large variations in CH_4-emissions between the measurement locations. The average cumulated emissions in TD-peat and IP-soil were 52 and 0.2 kg CH_4-C ha^{-1}, respectively in 2014, and 170 and 0.6 kg CH_4-C ha^{-1} in 2015. In the IP-soil, there were periods with a net uptake of CH_4. High concentrations of CH_4 in the soil air right above the buried peat (up to 45 vol %) indicated

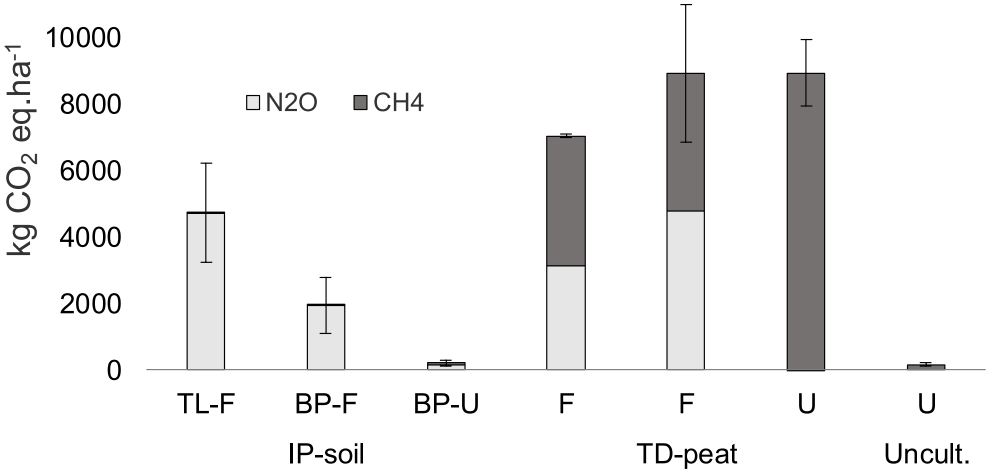

Figure 2. Cumulated greenhouse gas emissions from grassland on peat inversion (IP-soil) and tile-drained peat (TD-peat), and from an uncultivated peat the growing season 2015. The measurement locations on IP-soil are on the top of tilted mineral layer (TL), on the top of buried peat (BP), either fertilized (F) or unfertilized (U), and on the TD-peat, either fertilized (F) (two locations) or unfertilized (U). Emissions of N_2O and CH_4 are calculated as CO_2 equivalents per ha. The values are means of two parallel chambers at each location, with min and max levels indicated by the vertical line.

substantial CH_4 production in the buried peat, but only a minor part of the CH_4 reached the surface. This is probably because the mineral cover layer acted as an efficient scrubber for CH_4 by supporting growth and activity of microbial CH_4-oxidizers. In the uncultivated peat, the CH_4-emissions were small (4.1 kg CH_4-C ha^{-1} in 2015), but higher than in the IP-soil. Calculated as global warming potential (GWP) in CO_2 eq. per ha, the GWP based on CH_4 and N_2O emissions was higher in the TD-peat than in the IP-soil. This was most evident in 2015 (Figure 2).

Conclusions

Our case study suggests that inversion of earlier TD-peat can improve agronomic feasibility while reducing CH_4 emissions. More investigations, including C-flux measurements, are needed to prove that peat inversion reduces GHG emissions and slows down peat degradation relative to grassland cultivation on TD-peat drained peat.

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Effects of tractor traffic on soil compaction and grassland yield

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Abstract

A wetter future climate in Northern Europe may increase soil compaction by machinery traffic. We investigated the impact of tractor traffic on grassland yield, soil porosity and penetration resistance in three field trials on contrasting soils in different regions of Norway. The trials had a split-plot design with three levels of two wheel-by-wheel tractor passes after each cut (no traffic, light tractor, heavy tractor), three different seed mixtures and two fertilization levels. The yield reduction by tractor traffic was 26% at Løken, 4% at Fureneset and zero at Tjøtta. There was a positive correlation between yield reduction and the soil moisture content at wheeling. Tractor traffic reduced pore volume and air capacity and increased bulk density, degree of compactness and penetration resistance, with the largest effect on the silty soil at Løken and the smallest on the sandy soil at Tjøtta. There were no significant differences in either yield or soil physical properties between wheeling with light or heavy tractor. Soil texture and soil moisture content are thus major factors explaining traffic effects on soil physical properties and grassland yield.

Keywords: dry matter yield, penetration resistance, soil moisture, soil porosity, tractor weight

Introduction

The expected increased precipitation in Northern Europe will lead to wetter soils which are more susceptible to compaction by machinery traffic. Former Norwegian field experiments have concluded that traffic reduces grass yield, but the effect has differed a lot between trials and sites. The objective of this study was to investigate the effect of tractor traffic on grass yield, soil porosity and penetration resistance on contrasting soils in various climatic zones.

Materials and methods

Trials were located at Tjøtta in northern Norway (65°49'N, 12°25'E), Fureneset in western Norway (61°22'N, 5°24'E) and Løken in the east-central mountains (61°04'N, 09°04'E; 500 m a.s.l.) during 2011-2013. The soil at Tjøtta was medium sand (86% sand, 11% silt, 3% clay, 6% organic matter), at Fureneset silty medium sand (59% sand, 35% silt, 6% clay, 12% organic matter) and at Løken silt (14% sand, 81% silt, 5% clay, 5% organic matter). Normal precipitation and temperature (1961-1990) in the growing season (April-September) are 440 mm and 9.6 °C at Tjøtta, 835 mm and 10.5 °C at Fureneset and 335 mm and 8.5 °C at Løken. The precipitation was higher than normal at all sites in all three growing seasons, especially in 2011. The difference was greatest at Løken where the precipitation was 85, 46 and 68% higher than normal in the years 2011, 2012 and 2013, respectively.

Three levels of two wheel-by-wheel tractor passes were performed after each cut in 2011-2013: no traffic, traffic with a light tractor (3-4 Mg) and heavy tractor (6-7 Mg). These treatments had three replicates in each trial. Three seed mixes containing red (*Trifolium pratense* L.) and white clover (*Trifolium repens* L.) (0, 15 or 30% by weight) and a mixture of timothy (*Phleum pratense* L.), meadow fescue (*Festuca pratensis* L.) and smooth meadow-grass (*Poa pratensis* L.) were also tested, as well as two levels of fertiliser: 110 kg total N ha^{-1} as cattle slurry applied in spring and 170 kg N ha^{-1} including 60 kg N ha^{-1} from mineral fertiliser applied after the first cut.

Soil moisture content at wheeling was measured gravimetrically in samples taken at 10 cm depth intervals on each tractor traffic plot. Penetration resistance was measured after wheeling using Field Scout SC900 with a 60° cone of 1.26 cm^2 cross section on all small plots at 2.5 cm intervals to 30 cm. Undisturbed soil samples were taken in the autumn 2012 (Tjøtta), in 2012 and 2013 (Fureneset) and in 2013 (Løken). Two 100 ml cores were taken from selected plots at each of the following depths: 5-9 cm, 15-19 cm, 23-27 cm, except at Fureneset in 2012 when the deepest horizon was excluded. Pore size distribution, water storage, aeration properties, bulk density and the relative degree of compactness were measured in the laboratory as described by Riley (1996).

The field trials were cut twice a year and dry matter yield (DMY) was recorded for three ley years (2011-2013). Analysis of variance was applied separately to data from each site using split-plot models to evaluate the significance of tractor traffic, N fertilisation level and seed mixture on DMY and penetration resistance. Results are only presented here for the former. Analyses of variance of soil porosity were performed for each trial and for the combined data.

Results and discussion

There were significant differences between sites in most of the measured soil parameters. Air permeability was lowest and the relative degree of compactness was highest in the silt soil at Løken. Within sites, traffic tended to reduce total porosity, air capacity and air permeability and to increase bulk density and the relative degree of compactness (some results are shown in Table 1). The effects were however not significant in individual trials, due to few degrees of freedom. The combined results of all sites showed significant effects of traffic on total porosity, air capacity and relative degree of compactness in the two upper soil depths. The effect on bulk density was significant only at 5-9 cm and that on air permeability at no depth.

The effect of tractor traffic on DMY differed between sites (Table 2). At Løken it reduced DMY significantly (26% over three years), whereas at Fureneset the reduction was significant only at the second cuts and at Tjøtta no reduction was found. Use of a light versus a heavy tractor did not affect DMY differently in any of the trials.

When the soil moisture at wheelings made just after the first cut was expressed as a percentage of that held at field capacity, we found significant positive correlations between these values and the yield reductions

Table 1. Effects of tractor traffic on air capacity, air permeability and the relative degree of compactness at three depths. For Fureneset, mean values from 2012 and 2013 are shown.

	Tjøtta (2012			Fureneset (2012/2013)			Løken (2013)		
	No traffic	Light tractor	Heavy tractor	No traffic	Light tractor	Heavy tractor	No traffic	Light tractor	Heavy tractor
Air capacity (%)									
5-9 cm	23.4	22.7	21.4	8.3	8.2	6.4	11.9	5.8	6.9
15-15 cm	20.7	22.1	23.0	8.3	7.9	6.9	15.4	6.4	6.9
23-27 cm	25.2	23.5	23.5	9.3	9.3	9.7	11.3	7.5	7.0
Air permeability (μm^2)									
5-9 cm	33.5	34.5	33.3	11.5	6.0	7.2	6.5	1.9	0.4
15-15 cm	26.2	31.7	28.0	7.8	4.4	6.5	6.3	1.7	3.2
23-27 cm	28.6	27.2	27.3	4.3	4.0	5.2	4.8	3.2	0.7
Relative degree of compactness (%)									
5-9 cm	67.0	68.1	69.1	85.6	86.7	90.2	91.0	105.4	101.3
15-15 cm	73.8	72.0	73.2	85.1	87.5	89.0	89.7	101.7	99.7
23-27 cm	71.0	75.7	77.2	86.8	86.5	88.2	94.4	102.7	102.5

Table 2. Effect of tractor traffic on dry matter yield (DMY) (t $ha^{-1}y^{-1}$) at three sites. DMY of first cut and total DMY are means of 2012-2013 and DMY of second cut is mean of 2011-2013 (all data are averages of different seed mixtures and fertilization levels).

	Tjøtta			Fureneset			Løken		
	1st cut	2nd cut	Total	1st cut	2nd cut	Total	1st cut	2nd cut	Total
No traffic	4.35	3.31	7.41	5.48	2.88	8.30	3.52	2.35	5.96
Light tractor	4.45	3.23	7.44	5.46	2.65	8.08	2.77	1.43	4.22
Heavy tractor	4.38	3.14	7.30	5.30	2.54	7.82	2.89	1.65	4.56
s.e. of mean	0.086	0.057	0.107	0.145	0.060	0.176	0.079	0.094	0.137
P light vs heavy	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	0.16	n.s.
P traffic vs no traffic	n.s.	0.16	n.s.	n.s.	0.02	0.18	0.002	0.002	<0.001

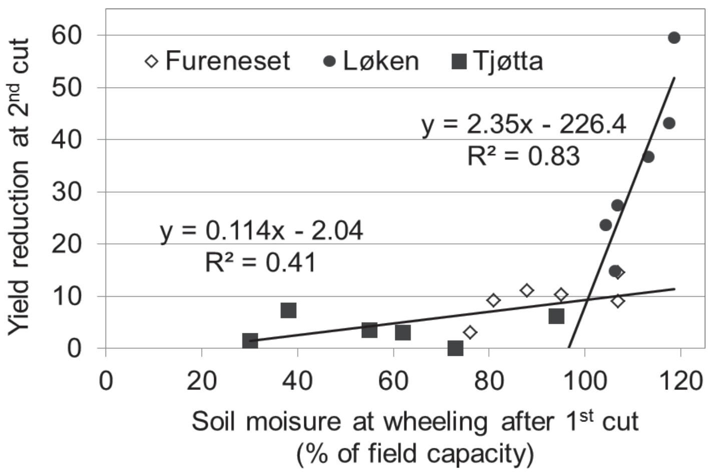

Figure1. Relationship between yield reduction (%) at second cuts and the soil moisture content at 0-10 cm depth, expressed as % of field capacity, at the wheelings performed after first cuts.

from tractor traffic at the second cuts (P=0.024 for Tjøtta and Fureneset together and P=0.011 for Løken). The yield reductions were greatest by far at Løken, where the soil was in all cases wetter than field capacity (Figure 1).

In autumn 2013, after six wheeling incidents, the soil mechanical resistance was significantly higher with traffic than without traffic at the depths 2.5-12.5 cm at Tjøtta, 2.5-17.5 at Fureneset and 2.5-22.5 cm at Løken (P<0.05). The effect was clearest at Løken were average penetration resistance through 0-30 cm depth was 1,489, 2,496 and 2,501 kPa in soil with no traffic, light tractor and heavy tractor, respectively. There were no significant differences between tractors of different weight at any of the sites.

Conclusions

Soil texture and soil moisture content at the time of wheeling are major factors which account for the variation in the effects of tractor traffic on soil physical properties and grassland yield.

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Rooting of permanent grassland in relation to build-up of soil organic matter for climate mitigation

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Abstract

Improved grassland rooting can have a positive effect on carbon sequestration. In our study we measured the root biomass and soil organic matter (SOM) of ten 'young' (<13 years) and ten 'old' (>18 years) grasslands on ten dairy farms on marine clay (fluvisol) in the north of the Netherlands. The aim of the study was to investigate the range of rooting in existing permanent grasslands in relation to grassland age and SOM build-up. The root biomass of the twenty permanent grasslands varied between 4.3 and 15.7 ton ash-free dry matter ha^{-1}. There was no significant difference in root biomass between old and young grasslands. SOM in the soil layer 0-10 cm was significantly higher in old grasslands and correlated positively with root biomass in 0-10 cm in old grasslands. By a theoretical calculation, we confirmed the possible long-term causality of measured differences in root biomass and SOM in old grasslands.

Keywords: permanent grassland, grass rooting, root biomass, soil organic matter, climate mitigation

Introduction

Higher grassland root biomass can result in a higher supply of organic matter to the soil (Abberton *et al.*, 2008). This increases carbon sequestration and can therefore be regarded as a tool for climate mitigation. There is a wide range in grassland root biomass which is related to e.g. grass species, cultivar and grassland management (Deru *et al.,* 2014). The aim of this study was to investigate the range of root biomass in existing young and old permanent grasslands on marine clay soils, the effect of grassland age ('young' vs 'old') and the relation between rooting and soil organic matter (SOM) build-up.

Materials and methods

On each of ten dairy farms on marine clay soil (fluvisol) in north of the Netherlands, two permanent grasslands were selected; a 'young' (<13 years) and an 'old' (>18 years) grassland. On each of these grasslands the root biomass was sampled in April 2014 at three soil depths (0-10; 11-20 and 21-30 cm) by taking three soil cores per plot per depth with a root auger (Ø 8.2 cm). The samples were thoroughly washed out with water over a sieve with a mesh size of 2 mm. All roots were collected (other non-root particles were removed by hand) and oven dried at 105 °C for dry matter measurement. After incineration at 600 °C, root biomass was corrected for percentage of ashes and expressed as ash-free dry matter (AFDM kg ha^{-1}).

SOM was measured in the soil layer 0-10 cm by taking a field-moist bulk sample of 70 cores (Ø 2.3 cm). A part of the bulk sample was oven-dried at 40 °C and SOM was determined by loss-on-ignition. An ANOVA procedure (Genstat 13.3, VSN international) to test for treatment effect ('young' versus 'old') on SOM and root biomass was used. Each of the 10 farms in which both treatments were compared was statistically regarded as a block. Pearson correlations between root biomass and SOM were calculated.

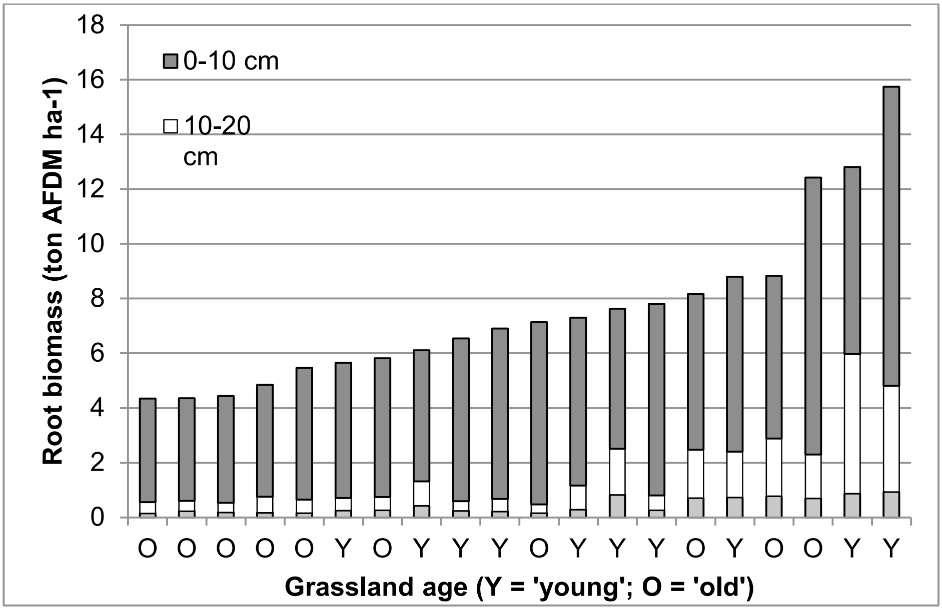

Figure 1. Range in root biomass ash-free dry matter (AFDM) in soil layer 0-30 cm (ton ha^{-1}) across the 10 'young' (<13 years) and 10 'old' (>18 years) grasslands.

Results and discussion

Mean root biomass in the soil layer 0-30 cm of the twenty grasslands was 7.6 t AFDM ha^{-1} and ranged from 4.3 to 15.7 t AFDM ha^{-1} (Figure 1). This is much higher than the 2.1 to 4.1 t dry matter ha^{-1} found by Deru *et al.* (2014) on sandy soil in the soil layer 0-24 cm. There was no significant difference between old and young grasslands in root biomass. In general root biomass increases with increasing grassland age up to 15 years (Whitehead *et al.*, 1990), however, van Eekeren *et al.* (2008) showed that root counts at 10 cm soil depth were 77% lower for 38 year-old grassland compared to three-year-old grassland.

SOM in the soil layer 0-10 cm was significantly (P<0.001) higher in old grasslands (13.3 g 100 g^{-1}) compared to young grasslands (10.7 g 100 g^{-1}). For old grasslands there was a significant (R^2 = 0.6; P<0.01) correlation between root biomass at 0-10 cm soil depth and SOM at 0-10 cm soil depth. The equation showed that a 1 ton increase in root biomass (AFDM) was associated with a SOM increase of 0.88% at 0-10 cm soil depth. We verified this observation by a theoretical calculation of long-term effects of a high versus low root biomass on SOM, using our experimental data. We assumed a fixed root turnover of 76% per year (Scott *et al,* 2012), a contribution of dead root material to build-up of SOM of 50%, a SOM decomposition of 2% per year, a soil bulk density of 1.03 g cm^{-3} and no difference in organic matter contribution from stubbles and leaves. With these assumptions, a difference in root biomass of 1 t ha^{-1} contributes in the long run to a difference in SOM of 1.8% at 0-10 cm soil depth. This calculated theoretical effect is larger than our observed effect, which may be partly due to the fact that the equilibrium state has not yet been reached in our grasslands.

In contrast to the old grasslands, we found no correlation between root biomass (AFDM) and SOM in the young grasslands (Figure 2). SOM in permanent grassland soils is the resultant of many years of organic matter accumulation from dying roots and other unharvested plant parts and input of organic manures. In young grasslands this process has recently been disturbed by soil tillage, resulting in mixing

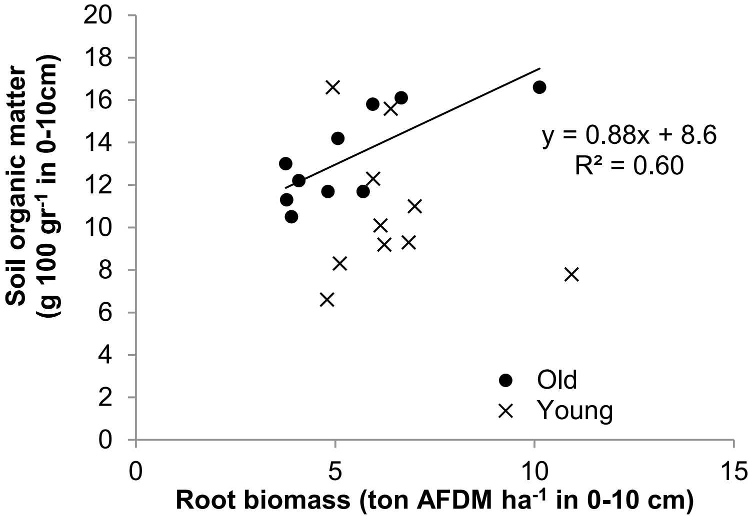

Figure 2. Relation between root biomass ash-free dry matter (AFDM) and soil organic matter in 'old'(>18 years) and 'young' (<13 years) grasslands, with linear regression line for 'old' grassland ($R^2 = 0.60$, P=0.009).

of organic matter into deeper soil layers and increased mineralisation in combination with a different cropping (arable) history.

Conclusions

There is a high variation of root biomass (AFDM) of permanent grasslands on marine clay soil. We did not find a significant difference between old and young grasslands in root biomass. In old (>18 years) grasslands a correlation between SOM and root biomass suggests that every ton root biomass (AFDM) results in an increase of 0.9% SOM at 0-10 cm soil depth. This can be theoretically justified by a calculation of long-term effects of a high versus a low root biomass on SOM.

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Analysis of changing climate impact on timothy productivity in two contrasting geographical locations

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Abstract

The aims of this study were: (1) to assess the trends of climatic variables in two contrasting geographical locations: central Poland and northern Norway; and (2) to evaluate the influence of the detected trends on timothy yields. This grass species was selected for its high importance for forage production in Norway as well as in Poland. For the assessment of climate trends, historical meteorological data, which cover time series from 1985 onwards, were used. Trends of various climate condition indicators were investigated. Data on timothy yields were collected beginning in the 1990s for Brody in Poland from cultivar testing experiments and Holt in Norway by the national cultivar-testing program. The results indicated that in central Poland air temperature in specific months significantly decrease the annual yield of timothy while in northern Norway many climatic variables, such as earlier start and prolonged length of growing season, may have a slightly positive impact on timothy productivity.

Keywords: climate change, grassland, productivity, timothy, yield, climatic trends

Introduction

The observed changes of climate conditions in recent decades vary depending on the region of Europe. Climate change affects grassland productivity in Poland as well as in northern Norway. In Poland, the most important abiotic factor limiting grassland productivity is water shortage and precipitation distribution during the growing season (Dąbrowska-Zielińska *et al.*, 2015). In northern latitudes (in Norway), the short growing season and harsh winters limit productivity. The winters are now becoming warmer, and extreme winter warming events may lead to de-hardening of grasses or more problems with ice encasement, thus increasing the risk of winterkill (Jørgensen *et al.*, 2010). The negative impacts of climate change on grassland yield can be mitigated by selecting grass species and cultivars that are best adapted to unfavourable weather conditions as components for seed mixtures. This justifies the need for investigating the impact of climate change on the yields of timothy, a very important grass species for forage production in Poland and Norway.

This study aims to evaluate the trends of meteorological variables in two locations in Europe, which greatly differ in terms of latitude and geography: central Poland and northern Norway, and to assess the influence of the detected trends on timothy yields.

Materials and methods

The meteorological data for the analysis were collected from 1985-2014 at the Experimental Station of Poznan University of Life Sciences (PULS) (52°26' N, 16°18' E; 92.0 m a.s.l.) located in Brody, central Poland and from 1989-2015 at Holt (69°39' N, 18°54' E; 20 m a.s.l.) located in Tromsø, northern Norway. The first stage of the analysis was searching for significant trends of climatic variables, which may have the greatest impact on grass biomass production. Using the Mann-Kendall test, trends of the following parameters were examined: average daily air temperature (annual and monthly), the start (first spell of seven days with temperatures above 5 °C in the year) and length of growing season (calculated until the end of the last spell of at least seven days with temperatures above 5 °C in the year), growing degree-days (0 and 5 °C base), and precipitation sums (annual and monthly). In the second stage of the analysis, simple linear regressions were run using timothy (*Phleum pratense* L.) dry matter (DM) yield as the dependent variable, and each of the climatic variables that showed significant trends were run as independent variables. Only annual DM yield was considered because in Poland the standard cutting regime of this grass was three cuts and in Norway two cuts per year. Data on yields of timothy cultivars were collected from the 1990s for Brody from cultivar testing experiments, and for Holt by the national program for cultivar testing. The statistical calculations were made in the R software environment (R Core Team, 2013). For trend analyses the 'Kendall' library was applied (McLeod, 2011), the remaining analyses being performed using the 'stats' library.

Results and discussion

The results show significant positive trends over time, as determined by the Mann-Kendall test, in the average annual air temperature, and average temperatures for six individual months in Brody, while in Holt, only May temperatures were found to be increasing with statistical significance (Table 1). Precipitation has decreased in December in Brody during the time period, whereas spring precipitation (March and April) has increased at Holt. Growing degree-days (base 5 °C) have increased in Brody and at Holt the same trend was determined in the case of growing degree-days but at base 0 °C. Growth start in spring is earlier, with an extended growing season at Holt. The results confirm the conclusions of other authors, e.g. Dąbrowska-Zielińska *et al.* (2015) and Karlsen *et al.* (2009), concerning significant climate changes in central Poland and northern Norway.

Simple linear regressions showed significant negative relationships of air temperatures in July, September and November on timothy DM yields in Brody. Higher air temperatures may limit productivity (Dąbrowska-Zielińska *et al.*, 2015) in vegetation season (water deficit) and also during winter by e.g. fungal infection, influencing mainly first regrowth in the following year. At Holt, there were significant positive impacts of May temperature, April precipitation, earlier start of growing season and length of growing season on timothy DM yields, but their explanatory powers were low, when considering R^2 (Table 2).

Table 1. Climatic variables with significant trends in Brody and Holt.[1]

Climatic variables	Kendall's tau statistic	Two-sided *P*-value	Kendall Score (S)	Denominator (D)	Variance of Kendall score
Brody					
Average air temperature yearly	0.34	0.0083**	149	435.0	3142
Average air temperature in April	0.42	0.0015**	178	425.4	3115
Average air temperature in June	0.36	0.0059**	155	431.0	3133
Average air temperature in July	0.32	0.0152*	137	433.0	3138
Average air temperature in August	0.30	0.0232*	128	430.5	3132
Average air temperature in September	0.30	0.0223*	129	432.0	3136
Average air temperature in November	0.30	0.0213*	130	433.5	3139
Sum of precipitation in December	-0.26	0.0497*	-111	435.0	3142
Growing degree-days (5 °C base)	0.38	0.0034**	165	435.0	3142
Holt					
Average air temperature in May	0.34	0.0124*	121	351.0	2301
Sum of precipitation in March	0.27	0.0551	88	324.5	2057
Sum of precipitation in April	0.29	0.0390*	100	350.5	2300
Start of growing season	-0.29	0.0403*	-99	343.9	2284
Growing degree-days (0 °C base)	0.35	0.0110*	123	351.0	2301
Length of growing season	0.40	0.0042**	138	344.4	2286

[1] *, **, *** denote significance at the 0.05; 0.01; 0.001 levels, respectively.

Table 2. Impact of climatic variables showing significant trends on the annual DM yield of timothy in Brody and Holt.[1]

Climatic variables	Regression coefficient	Standard error	t value	Significance of the relationship (*P*-value)	Adjusted R squared
Brody					
Average air temperature yearly	0.003	0.0050	0.68	0.49874	-0.003
Average air temperature in April	-0.124	0.1542	-0.80	0.42217	-0.002
Average air temperature in June	-0.010	0.0908	-0.12	0.90853	-0.005
Average air temperature in July	-0.579	0.0662	-8.74	0.00000***	0.259
Average air temperature in August	0.112	0.0639	1.75	0.08112	0.009
Average air temperature in September	-0.825	0.1022	-8.08	0.00000***	0.229
Average air temperature in November	-0.312	0.0802	-3.88	0.00014***	0.061
Sum of precipitation in December	0.108	0.0849	1.27	0.20427	0.003
Growing degree-days (5 °C base)	0.001	0.0006	1.37	0.17217	0.004
Holt					
Average air temperature in May	0.007	0.0023	2.85	0.00466**	0.017
Sum of precipitation in March	-0.002	0.0039	-0.59	0.55681	-0.002
Sum of precipitation in April	0.226	0.0894	2.52	0.01201*	0.013
Start of growing season	-0.076	0.0113	-6.76	0.00000***	0.097
Growing degree-days (0 °C base)	0.001	0.0009	0.82	0.41316	-0.001
Length of growing season	0.005	0.0012	4.60	0.00001***	0.046

[1]*; **; *** denotes significance at the 0.05; 0.01; 0.001 levels, respectively.

Conclusions

In two contrasting geographical locations in Europe, the analysed climatic variables showing significant trends over recent decades were different. Among them, significant negative effects on DM yield of timothy were confirmed for increased air temperatures in July, September and November in central Poland, while a positive, but hardly relevant impact on yields was found for increased air temperature in May, April precipitation, and earlier start and prolonged length of growing season in northern Norway.

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The degree of perenniality of timothy (*Phleum pratense* L.) accessions is related to geographic origin

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Abstract

Timothy (*Phleum pratense* L.) is a commonly grown perennial forage grass in temperate areas. Since stem formation and flowering tillers contribute significantly to canopy structure and eventually to herbage biomass accumulation, the regulation of tiller development is of interest. Perennials grown at high latitudes require different growth and flowering strategies than annuals in order to tolerate several growth cycles under harsh conditions. It has been shown that perennial species, e.g. *Arabidopsis lyrata*, have stronger apical dominance, and thus higher degree of perenniality. A greenhouse experiment was conducted to determine differences in growth strategies between timothy accessions of different origin after vernalization durations. Results showed that prolonged vernalization increased the number of flowering tillers in all the studied accessions. Northern accessions produced more vegetative tillers and showed a higher degree of apical dominance. It seems that timothy might have some similarities to *A. lyrata* in the processes determining degree of perenniality. Lateral tillers possibly maintain perenniality and this can represent the more perennial growth habit of northern accessions, and adaptation to shorter growth period at high latitudes. It can be speculated that the geographic origin of timothy accessions is also related to the degree of perenniality.

Keywords: apical dominance, perenniality, *Phleum pratense* L., tiller type, timothy, vernalization

Introduction

Timothy (*Phleum pratense* L.) is a perennial forage grass that can persist in grass-mixtures for up to five years. Perennial species grown at high latitudes must have specific growth and flowering strategies, compared with annuals, in order to tolerate several growth cycles under harsh conditions. For hardy perennials it is typical that not all apices will develop into generative apices, and part of the meristems are saved for growth in subsequent seasons (Wang *et al.*, 2009). Some plant species have the ability to 'remember' vernalization even after a long period of time, and this ability or lack of it determines the potential for perennial growth habit (Woods *et al.*, 2014). In addition, it has been shown that perennial species, e.g. *Arabidopsis lyrata* populations, have strong apical dominance, and thus a higher degree of perenniality (Remington *et al.*, 2015). The lateral meristems of forage grasses do not have a real endodormancy but rather the trait has been associated with a degree of regrowth. In timothy breeding programmes regrowth after cutting is an important trait of interest, as well as the possibility to have cultivars with longer life cycle. The aim of this study was to investigate the growth rhythm and perenniality of different timothy accessions during and after vernalization treatments.

Materials and methods

In a greenhouse experiment the effect of vernalization duration on the developmental rate and formation of different tiller types was studied in eleven timothy accessions of distinct geographic origin. In this paper results are focused on the most extreme accessions, BOR S (southern origin, no vernalization requirement for flowering) and BOR N (northern origin, obligatory vernalization requirement for flowering) (Jokela *et al.*, 2015). Clonal material was propagated from lateral tillers, and the tillers were first grown for four weeks at 12-h DL (20/15 °C day/night) in fertilized and limed peat (Kekkilä B2, Finland). Plants were transferred for vernalization (0, 2, 10, 12 or 15 weeks) at 6/4 °C (day/ night) 8-h

DL in growth chambers (Weiss Technik, Germany) arranged in a completely randomized design with three replications. Non-vernalized plants were grown at 12-h DL (20/15 °C day/night) for four weeks. After vernalization all plants were transferred to 16 h DL (20/15 °C day/night) greenhouse conditions. Tillers produced during the vernalization or SD conditions were marked with rubber bands immediately after these treatments. The formation of different tiller types (GEN, ELONG, VEG) (Virkajärvi *et al.*, 2012) during and after vernalization in main and lateral tillers was reported at the end of the experiment, after ca. six weeks growth. Results were analysed (ANOVA) using IBM SPSS software (Version 23), replicates as random factors and vernalization and accessions as fixed factors.

Results and discussion

To reveal the effect of vernalization time on the formation of lateral tillers, a total of eleven distinct timothy accessions were studied. ANOVA showed significant differences between tested accessions (P<0.0001) but not between vernalization treatments (P = 0.165) in the total number of tillers produced. Detailed analysis on the most southern accession (BOR S) showed its ability to produce more GEN lateral tillers compared with the northern BOR N (Figure 1). In contrast, BOR S had significantly fewer VEG tillers than BOR N (Figure 1). In perennial *Arabidopsis lyrata* the degree of perenniality is defined partly by the number of VEG and GEN tillers, so that populations with a high degree of perenniality have more VEG tillers and fewer GEN tillers (Remington *et al.*, 2015).

Results showed that prolonged vernalization increased the number of GEN main and lateral tillers in all studied accessions (Figure 1). Vernalization saturation progressed stepwise from main to lateral tillers, and was more synchronized in southern accessions. In *A. lyrata* it was reported that delay in the transition to reproductive stage and higher level of apical dominance resulted in reduced number of GEN tillers, and thus in a higher degree of perenniality (Remington *et al.*, 2015).

For *A. lyrata* Remington *et al.* (2015) had hypothesized that a more perennial population would have earlier shoot initiation, but this was not seen in their study. In our study, however, BOR N produced more tillers during vernalization, prior to transition to flowering (Figure 1a), possibly indicating higher degree of perenniality according to the original hypothesis of Remington *et al.* (2015). In addition, in

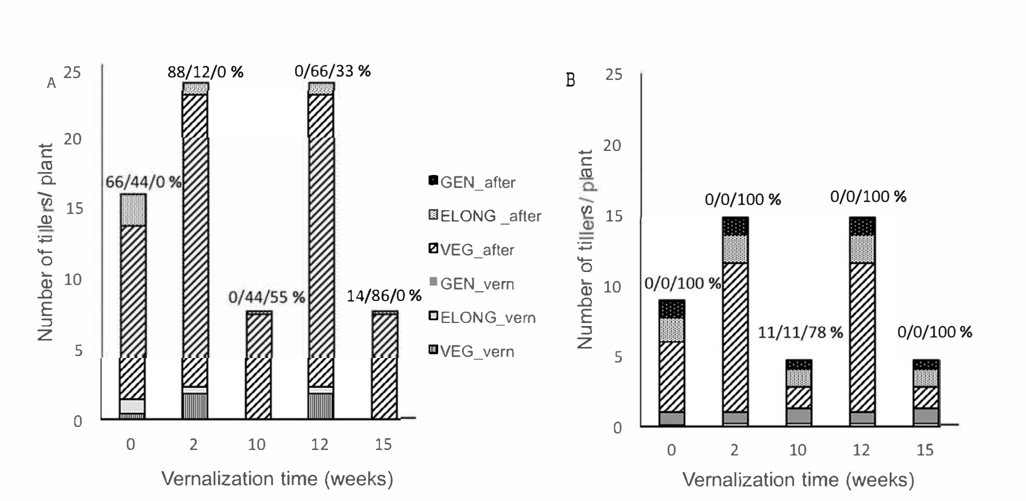

Figure 1. The number of tiller types per plant (VEG, ELONG, GEN) in lateral tillers during and after five different vernalization durations (0, 2, 10, 12 or 15 weeks) and six weeks growth at 16h DL in (A) BOR N and (B) BOR S, n=9. Percentage values on the top of the bars represent the proportion of the main tiller type VEG/ELONG/GEN in each vernalization treatment.

perennial *Arabis alpina*, shoots produced after vernalization remained vegetative (Wang *et al.*, 2009), and this was also seen in BOR N, where most of the tillers produced after vernalization were VEG tillers (Figure 1a). In field conditions we have seen that northern accessions have poorer regrowth ability (unpub. data), which might partly be due to higher degree of perenniality in northern accessions, because of their inability to 'remember' vernalization as proposed by Woods *et al.* (2014).

It seems that timothy and *A. lyrata* might have some similarities in the processes determining the degree of perenniality. Lateral tillers possibly maintain perenniality and this can represent the more perennial growth habit of northern accessions, and adaptation to shorter growth periods at high latitudes. It can be speculated that the geographic origin of timothy accessions is also related to their degree of perenniality.

Conclusions

It seems that there is some similarity between timothy and *A. lyrata* in terms of processes regulating perenniality, and that northern accessions have higher degrees of perenniality than southern ones. Fast and synchronized development of GEN tillers was a characteristic of southern accession. In leys harvested for silage these characteristics can be seen as rapid regrowth and uniform development of tillers, and thus in a rapid decrease in digestibility. In timothy breeding programmes high yielding southern and high quality northern germplasm is combined. These results highlight that perenniality in timothy can be translated to different rates of tiller development, and thus, to different quantity and quality of herbage biomass.

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Incidence of root pathogens associated to clover root rot in Sweden

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Abstract

Red clover is an important forage legume, but its persistence in Swedish leys is limited by root rot associated with several soil-borne plant pathogens. In this study DNA of the pathogens associated to clover root rot was quantified using real-time PCR. A field experiment was set up at an organic farm in central Sweden and eleven future- and market cultivars were sown. Pathogen identification and quantification by real-time PCR was performed in roots of three selected cultivars sampled at different time points the seeding year. Internal disease severity indices were determined by visual assessment. *Fusarium avenaceum*, *Phoma* spp. and *Cylindrocarpon destructans* were identified in the analysed roots on the first sampling occasion. Significant differences in the amount of detected pathogen DNA were found within the sampling year for each of the pathogens. *Fusarium culmorum* appeared sporadically.

Keywords: red clover (*Trifolium pratense*), root rot, qPCR

Introduction

Red clover (*Trifolium pratense*) is an important forage legume in Sweden. Yield and nitrogen fixation may be severely reduced over time due to infection by soil-borne fungal pathogens causing root rot. Although it is a perennial species the plants rarely persist more than two harvest years (Wallenhammar *et al.*, 2008). Extensive surveys in Sweden have shown that red clover plants show symptoms of root rot already in the year of planting. The fungal pathogens causing red clover root rot in Sweden have been investigated previously, and mainly include *Fusarium avenaceum, Fusarium culmorum*, *Cylindrocarpon destructans* and *Phoma* spp. (Rufelt, 1986; Lager and Gerhardsson, 2002; Wessén, 2006, Öhberg, 2008).

For identification of soil-borne pathogens causing disease symptoms in roots, fast and specific diagnostic methods are needed. Quantitative real-time PCR (qPCR) is an objective, specific and reproducible technique appropriate for pathogen diagnostics and quantification. The aim was to identify the complex of soil-borne pathogens associated to root rot and to monitor disease development in field-infected red clover roots by qPCR.

Materials and methods

A total of 11 red clover cultivars were evaluated in a field experiment in central Sweden established on 29 April 2011. The trial was set up using a randomized block design with four replicates (each measuring 4.5×10 m), thus a total of 44 plots were sampled. Visual assessment of root rot was performed in all cultivars according to Rufelt (1986) and a disease severity index calculated. Three cultivars were selected for qPCR analysis; SW Torun, SW Vivi and SW Yngve. Roots from these cultivars were sampled on five occasions during the sowing year; 30 June, 28 July, 23 August, 22 September, and 10 November. Roots collected from the same plot on the same time point were treated as one root sample (2-25 roots). Each sample was cut into pieces by a pair of sterile scissors or mixed using a Retsch Grindomix. DNA was extracted using the FastDNA SPIN Kit (MP Biomedicals) according to a modified protocol. The following pathogens were amplified and target sequences detected and quantified from *F. avenaceum, F. culmorum, Phoma* spp. and *C. destructans* and *T. pratense.* Information on extraction methods, primers and probes, and qPCR analysis are described in Almquist (2016).

Results

DNA from *F. avenaceum, Phoma* spp. and *C. destructans* was detected on all sampling occasions (Figures 1A-C). Significant differences in the amount of detected pathogen DNA were found within the sampling year for each of the pathogens.

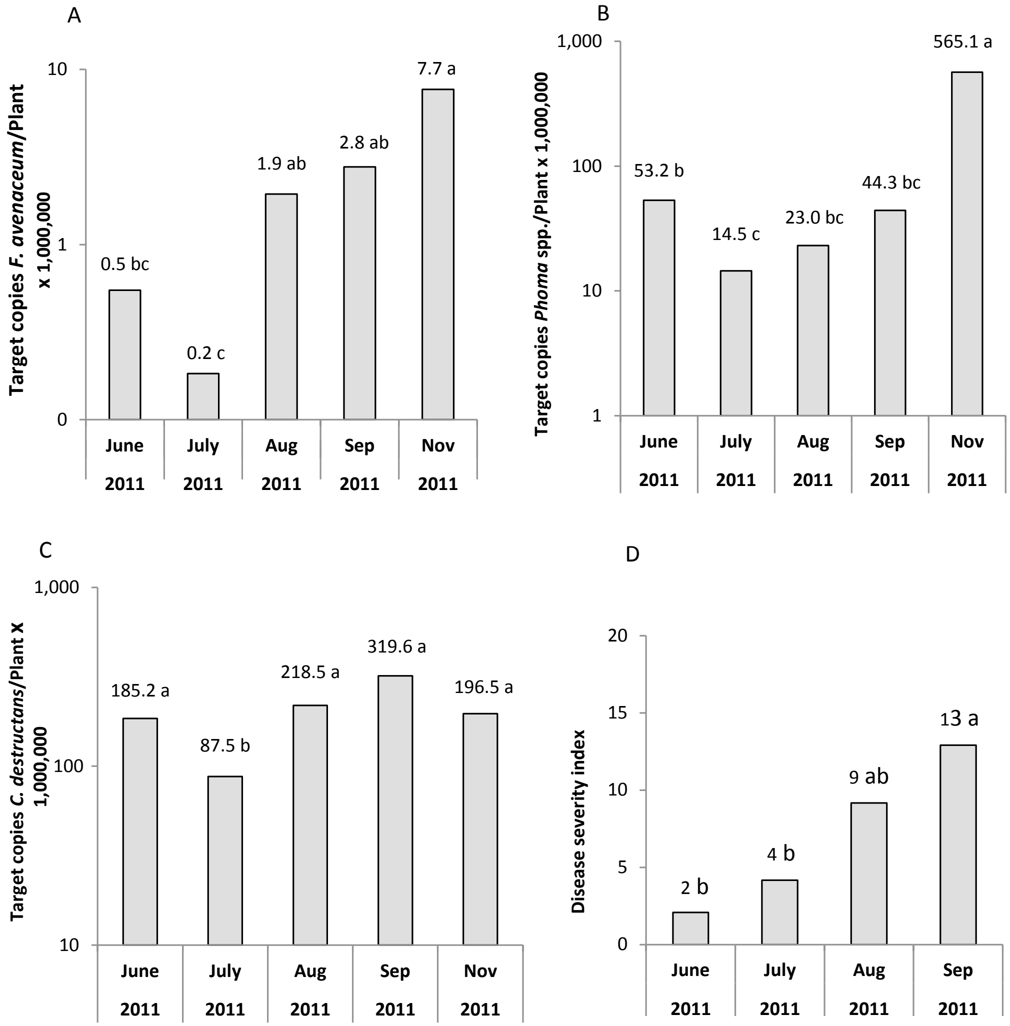

Figure 1. Disease progress over time demonstrated as the amount of DNA of (A) *Fusarium avenaceum*, (B) *Phoma* spp. and (C) *Cylindrocarpon destructans* (D) Internal disease severity indices (DSIs). All bars represent the mean of the red clover cultivars SW Torun, SW Vivi and SW Yngve collected from 4 different plots (n=12). Different letters indicate statistically significant differences between time points of sampling according to Tukey's HSD test (P<0.001).

The highest levels of *F. avenaceum* and *Phoma* spp. DNA were found in the roots sampled on the last sampling occasion (Figures 1A and 1B), while *C. destructans* was at a similar level throughout the season (Figure 1C). Visual assessments showed a significant increase in disease severity from July to September (Figure 1D). *F. culmorum* appeared sporadically.

Discussion

In this study, we monitored the presence of *Phoma* spp, *C. destructans* and two selected *Fusarium* species; *F. avenaceum* and *F. culmorum*, the seeding year of red clover. The pathogens occurred in the roots early in plant development, and three of the pathogens were detected already on the first sampling occasion in June, eight weeks after sowing, although most roots lacked visible symptoms. Thus, in addition to being a useful technique for disease identification in roots, qPCR can serve as an effective tool for pre-symptomatic pathogen detection in seedlings. *F. culmorum* was only detected in a few single samples at low levels which is consistent with previous studies where *F. culmorum* rarely caused severe symptoms (Ylimäki, 1967; Lager and Gerhardsson, 2002). Further studies are now required to screen the red clover roots for other potential pathogens contributing to the disease symptoms.

Conclusions

In summary, we have demonstrated that qPCR provides a useful tool to monitor the pathogens causing disease symptoms and is a tool for the first step towards a disease risk assessment of red clover root rot.

Acknowledgements

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Genome-wide allelic shifts in forage crops when grown at five diverse locations across Norway

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Abstract

The current germplasm of forage crops like perennial ryegrass (*Lolium perenne* L.), timothy (*Phleum pratense* L.) and red clover (*Trifolium pratense* L.) are not adequately adapted to future climatic conditions at higher latitudes. The climate is predicted to be more unstable during winter, and winter survival needs to be improved. In this study, the aim was to detect the effects of selection/local adaptation by estimating genome-wide shifts in allelic composition of single nucleotide polymorphism markers in samples from swards of perennial ryegrass, timothy and red clover grown and managed at diverse locations in Norway. In addition separate mixtures of cultivars and breeding populations of perennial ryegrass, timothy and red clover was sown at five geographically distinct locations throughout Norway. The fields were harvested for 3 years and leaf tissues sampled randomly from about 200 plants field^{-1} year^{-1}. To detect allelic shifts, genotyping by sequencing was used to generate genome-wide allele frequency fingerprints (GWAFFs). These allele frequency fingerprints were used to monitor shifts in population structure in response to location and years. Preliminary analyses demonstrate that the GWAFFs clearly distinguished samples from years/fields with good survival from those with poor survival based on scoring of winter survival in spring in perennial ryegrass and red clover.

Keywords: *Lolium perenne, Phleum pratense, Trifolium pratense,* single nucleotide polymorphism, genome-wide-allele-frequency-fingerprints, genotyping-by-sequencing

Introduction

The predicted climate change with higher temperatures and longer growing seasons will make it possible to extend the cultivation of certain forage crops like perennial ryegrass further north in Northern Europe. The existing genetic variation in most forage crops is neither wide enough nor adequately adapted to specific climatic conditions and management purposes to cope with the predicted future climate. It will require plant types which respond to an extended growth season by an early growth start without compromising tolerance to late winter/spring frosts, and which sustain more cuts and cease growth in time for winter survival. Our aim was to find out whether genome-wide shifts in allelic composition could be detected and used to identify genomic regions and candidate genes involved in local adaptation, which might be used in breeding locally adapted cultivars.

Materials and methods

Plant material used in this study were an equal seed-mixture of five diverse diploid populations of perennial ryegrass (two European cultivars (Arsenal and Toronto), one Norwegian cultivar (Fagerlin), and two Norwegian breeding populations (FuRa0575-79 and FuRa9805)), an equal seed mixture of MRL 95-3, MRL 97-2, LøRk0288 diploid red clover; and an equal seed mixture of MTL9701 + Grindstad timothy. These three forage crop mixtures were sown at five geographically distant locations throughout Norway, i.e. Bjørke (60°47'N), Særheim (58°45'N), Kvithamar (63°29'N), Bodø (67°28N) and Alta (69°58'N). The fields were harvested for 3 years and leaf tissues sampled randomly from ca.

200 plants field^{-1} year^{-1}. Winter survival was recorded as percentage coverage on the plots in spring of 2011, 2012 and 2013. To detect allelic shifts separate pools of leaf samples from 200 plants plot^{-1} were established for each location and year. DNA was extracted and genotyping by sequencing (GBS) was used to generate genome-wide allele frequency fingerprints (GWAFFs) for each pool (Elshire *et al.,* 2011). The UNEAK pipeline was used for GBS data analysis (Lu *et al.,* 2013), and allele frequencies were determined using the GWAFF procedure (Byrne *et al.,* 2013). PCA analysis was performed by the GAPIT program in R (Lipka *et al.,* 2012).

Results and discussion

Winter survival of perennial ryegrass, timothy and red clover was recorded in 2011, 2012 and 2013 after establishing the initial populations in 2010. Perennial ryegrass has good winter survival across three years at Bjørke, Kvithamar and Særheim but a very poor survival rate at the northernmost locations Alta and Bodø. Timothy has very good winter survival across three years at Bjørke and Kvithamar but relatively poor winter survival was observed at Alta, Bodø and Særheim in the final year 2013. Red clover has good winter survival at Bjørke, Kvithamar and Bodø, while poor survival was observed at Alta and Særheim. Overall, the locations show large differences in winter survival, with highest winter survival observed at Bjørke and Kvithamar, and lowest survival, as expected, at the northernmost location Alta for these species.

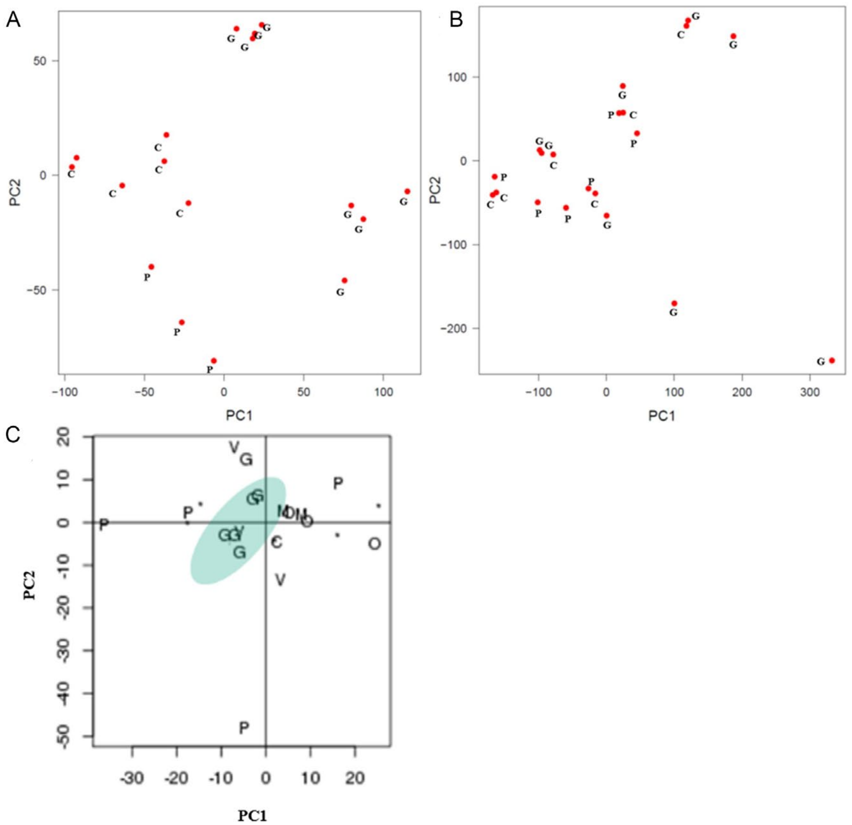

Figure 1. Distinguishing winter-survival ability groups based on GWAFFs in (A) red clover (B) timothy and (C) perennial ryegrass. Groups denoted in PCA plots as *C*: original population sown in 2010; *G*: Good winter survival; and *P*: Poor winter survival. In Figure 1C: perennial ryegrass has additional groups denoted as *V*: Very good winter survival. *O*: different combination mix of five populations (one mix is from two European cultivars Arsenal and Toronto; another mix from two Norwegian populations FuRa0575-79 and FuRa9805). Five dots (·) represent five individual populations (Arsenal, Toronto, Fagerlin, FuRa0575-79 and FuRa9805).

In perennial ryegrass, GBS was also performed in five individual populations (Arsenal, Toronto, Fagerlin, FuRa0575-79 and FuRa9805) and in different combination mix of five populations (one mix is from two European cultivars Arsenal and Toronto; another mix from two Norwegian populations FuRa0575-79 and FuRa9805) to track the allele frequencies detected in the three years in field. Allele frequencies were determined at 101,375 Single nucleotide polymorphism (SNP) positions using the GWAFF procedure (Byrne *et al.,* 2013). In order to assign an allele frequency to a SNP site in any sample, the read depth had to be at least 10. All SNPs have a 'minor allele frequency' of 5%, calculated by simply averaging the allele frequency calls. SNPs with more than 20% missing samples were eliminated. Similar filtering methods were employed in red clover at 32,854 SNP positions and in timothy at 222,974 SNPs. Further, the GWAFFs were used to monitor shifts in population structure in response to locations and years. PCA analyses of GWAFFs clearly distinguished samples from years/fields with good survival from those with poor survival based on scoring of winter survival in spring in red clover and perennial ryegrass (Figures 1A and 1C). However, samples from years/fields of timothy were not separated and this could be due to the small differences in winter survival across years at the locations (Figure 1B).

Conclusions

Our studies showed clear differences in winter survival within and among the species across the three years at five diverse locations in Norway. Highest winter survival was observed at Bjørke and Kvithamar, and lowest winter survival at Alta. GWAFFs clearly distinguished the good and poor winter survival locations and the genomic regions with significant shifts in allele frequency which might be important for local adaptation. Further work including the detection of F_{ST} outlier SNPs and annotation of the SNPs will reveal genes involved in selection and local adaptation, which can be useful in breeding locally adapted cultivars.

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Impacts of spring and summer droughts on yield and forage quality of three grasslands

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Abstract

An experiment was carried out on three semi-natural grasslands, characterized by contrasted climatic conditions, in order to explore the responses of herbage production and quality to drought. Drought was simulated under rain shelters, either in spring or in summer. Two factors were tested: intensity of use (intensive vs extensive) and watering regime (control vs drought). Drought reduced the dry matter production on average across sites and intensity of use by 12% during spring and by 52% in summer, without significant changes of the botanical composition. Regarding the forage quality, drought resulted in a slight decrease of crude protein content, but this response was uneven. Acid detergent fiber generally decreased, whereas water soluble carbohydrates increased. The concentrations of phosphorus were systematically lowered by water shortage. The interactive effects of drought and intensity of use were small. Although there was no consistent trend, drought impacted forage quality, but less than yield.

Keywords: rain shelter, permanent meadows, elevation gradient, nutrient content

Introduction

Drought affects forage production (yield and quality). The effects are, however, difficult to predict due to potentially very diverse environmental conditions. More specifically, the responses of grasslands to drought depend on the timing of stress (seasonality) and its severity. Management intensity is another factor that interacts with drought (Vogel *et al.*, 2012). The objective of this study was to investigate the forage responses of different grassland types to spring and summer droughts, in interaction with two management schemes. We hypothesised that: (1) the effects of drought are more pronounced on lowlands than highlands grasslands; and (2) intensive management amplifies the drought impacts, compared to the extensive management.

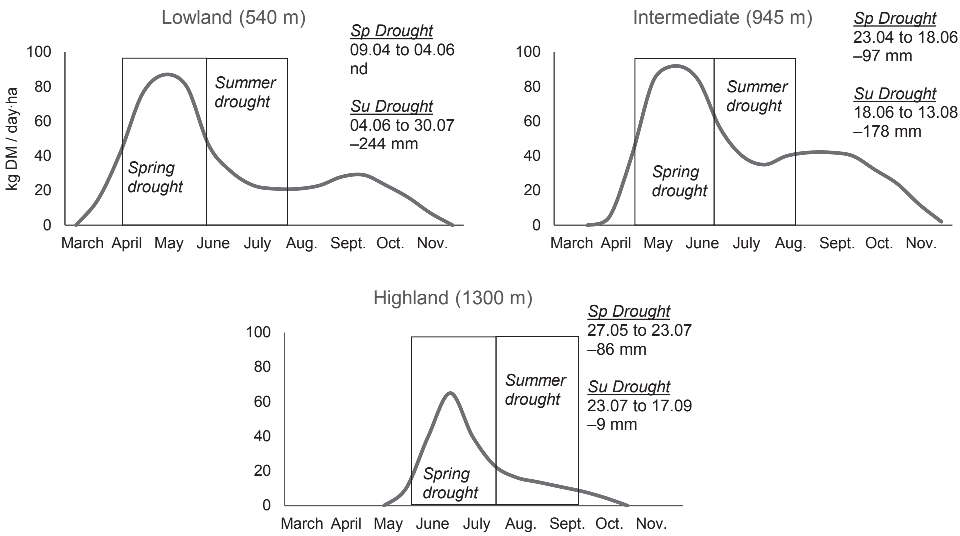

Figure 1. Timing of the drought treatments. Dates of spring (Sp) and summer (Su) droughts are given together with P – ETP at each site.

Materials and methods

The experiment was conducted in 2015 on three permanent meadows in the Swiss Jura mountains along an elevation gradient. The more productive sites at lower and intermediate elevation (lowland, 540 m; intermediate, 945 m) were dominated by *Lolium perenne*, *Trifolium repens*. The highest site (highland, 1,300 m) was mainly composed of *Festuca rubra* and *Agrostis capillaris*. A complete randomized block design with five replicates was set up in the three sites. Two factors were tested: watering regime (control vs drought) and intensity of use (intensive vs extensive). Drought was simulated under rain shelters, either in spring or in summer, during 8 weeks. The timing of the periods of stress was based on the seasonal dynamic of the growth (Figure 1). Control and drought plots were watered with 100%, respectively 30% of the 30-year precipitation average. The plots in the intensive management were cut at 4-week intervals, compared with 8-week intervals in the extensive management. Cutting dates were defined in order to harvest intensive and extensive plots simultaneously at the end of the drought periods. Botanical composition was determined according to the pin-point method of Daget and Poissonet (1971). The 4 m^2 plots were cut by means of a motor mower. In order to assess the effects of the treatments across the whole period of drought, the dry matter (DM) of the two cuts in the intensive management were compared with one cut in the extensive one. By extension, the data on forage quality in the intensive management were obtained by weighting the nutrient content of the two harvests by their relative yield

Table 1. Effect of spring drought, intensity of use and their interactive effect on dry matter yield and forage quality of the three grasslands (lowland, intermediate and highland).[1]

	intensive use		extensive use		*P*-value		
	control	drought	control	drought	watering	int. use	wr × int.
DM (Mg ha^{-1})							
lowland	4.09	3.62	5.74	5.86	0.973	<0.001	0.672
intermediate	5.30	4.36	7.32	5.91	0.002	<0.001	0.469
highland	3.85	3.65	4.01	3.82	0.157	0.209	0.968
CP in DM (g kg^{-1})							
lowland	152	159	89	88	0.360	<0.001	0.326
intermediate	157	146	88	86	0.083	<0.001	0.227
highland	145	138	111	106	0.045	<0.001	0.733
ADF in DM (g kg^{-1})							
lowland	284	277	333	325	0.233	<0.001	0.956
intermediate	273	269	345	319	0.014	<0.001	0.052
highland	270	270	323	318	0.342	<0.001	0.399
WSC in DM (g kg^{-1})							
lowland	135	141	167	187	0.303	0.006	0.575
intermediate	140	173	165	190	0.000	0.002	0.529
highland	175	192	127	140	0.009	<0.001	0.612
P in DM (g kg^{-1})							
lowland	3.2	3.2	1.8	1.7	0.636	<0.001	0.657
intermediate	4.0	3.6	2.7	2.2	<0.001	<0.001	0.620
highland	3.2	3.0	2.8	2.5	0.003	<0.001	0.481

[1] DM = dry matter; CP = crude protein; ADF = acid detergent fiber; WSC = water soluble carbohydrates; P = phosphorus.

Results and discussion

During the spring drought, forage production was solely impacted in the intermediate site (Table 1). The lowland site was hit end of April by heavy rainfall, so that there was no real stress effect (soil under the shelters became wet). In the highland site, the grass growth was sustained even with only 30% of the 'normal' precipitation. The relatively low evapotranspiration and, to a lesser extent, the clay soil could explain the absence of effect. Regarding forage quality, the values for the three sites were similar within each management intensity, in line with the comparable stage of maturity. Not surprisingly, the intensity of use had a strong influence on all parameters. Although to a lower extent than management, drought had an effect: crude protein (CP) and acid detergent fiber (ADF) tended to decrease; water soluble carbohydrates (WSC) increased and plant phosphorus (P) decreased.

There were no changes in the proportion of the main plant species at the end of the spring drought (data not shown), suggesting that the variations in forage quality caused by drought were mainly related to 'direct' effects on the plants (i.e. changes in chemical composition and morphology) but also to side effects on plant nutrition (e.g. lower soil NO_3^- concentrations under drought; data not shown).

The summer drought reduced DM yield by about 50% in the three sites (Table 2). The importance of the summer losses must be tempered by the fact that only a small part of the annual forage is produced

Table 2. Effect of summer drought, intensity of use and their interactive effect on dry matter yield and forage quality of the three grasslands (lowland, intermediate and highland).[1]

	intensive use		extensive use		*P*-value		
	control	drought	control	drought	watering	int. use	wr × int.
DM (Mg ha^{-1})							
lowland	1.45	0.81	1.06	0.58	<0.001	0.006	0.432
intermediate	3.51	2.32	2.11	0.33	<0.001	<0.001	0.050
highland	0.36	0.18	0.61	0.30	<0.001	0.001	0.123
CP in DM (g kg^{-1})							
lowland	159	157	128	128	0.783	<0.001	0.701
intermediate	195	182	164	178	0.899	0.001	0.007
highland	184	167	157	152	0.020	<0.001	0.188
ADF in DM (g kg^{-1})							
lowland	250	244	254	241	0.043	0.923	0.453
intermediate	254	253	286	225	<0.001	0.676	<0.001
highland	220	231	230	238	0.043	0.069	0.690
WSC in DM (g kg^{-1})							
lowland	129	144	155	169	0.042	0.002	0.955
intermediate	84	100	78	107	<0.001	0.905	0.114
highland	172	153	184	193	0.669	0.034	0.213
P in DM (g kg^{-1})							
lowland	3.0	2.6	2.2	1.8	0.006	<0.001	0.776
intermediate	5.2	4.9	3.7	2.3	<0.001	<0.001	<0.001
highland	3.7	3.3	3.5	3.0	0.002	0.051	0.702

[1] DM = dry matter; CP = crude protein; ADF = acid detergent fiber; WSC = water soluble carbohydrates; P = phosphorus.

during this period. There were no consistent effects of drought on CP and especially ADF. Interactive effects were observed in the intermediate site: responses to drought differed between the management schemes, with especially an important decrease of ADF in the extensive plots. Given the relative stability of NDF (data not shown), it can be assumed that the decrease of ADF has been counterbalanced by an increase of hemicellulose.

Conclusions

The relative reduction in yield caused by the drought was more pronounced in summer than in spring. Our results did not reveal a clear pattern of responses for nutrients, unless for WSC and P. Drought had the most impact on the intermediate site, whereas the site in the highlands was the less affected. This experiment suggests that drought has direct and indirect effects (i.e. on soil nutrient availability), interacting in a complex way on forage quality.

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Variability in germination under extreme temperatures of two perennial pasture legumes

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Abstract

Temperature is one of the most significant environmental factors affecting seed germination, and extreme temperatures can have detrimental effects on seed germination and seedling growth. Alfalfa (*Medicago sativa* L.) and sainfoin (*Onobrychis viciifolia* Scop.) are two perennial pasture legumes that hold an important place in cultivated grasslands. The objective of the study presented here was to analyse germination variability of these two species in response to a range of temperatures. The response of *M. sativa* showed statistically significant differences (P<0.01) between varieties. This was not the case for the two varieties of *O. viciifolia* tested. The findings of this study suggest that the germination of varieties of *M. sativa* were little affected by low temperature (5 °C) whereas germination at 40 °C was lower and showed high variability. On the other, hand, the two varieties of *O. viciifolia* were highly sensitive to extreme high (40 °C) temperature.

Keywords: *Medicago sativa*, *Onobrychis viciifolia*, genetic variability

Introduction

The annual time course of temperature is one of the most important factors affected by climate change. The International Panel on Climatic Changes (IPCC) anticipates an increase in global average and extreme temperature between 3.7 to 4.8 °C by 2100 (IPCC, 2014). Temperature will be subject to larger fluctuations between years, due to a higher frequency of extreme climatic events. Temperature is important in controlling seed germination. It is well documented that germination rate is highly temperature dependent (Bewely and Black, 1994). Alfalfa *Medicago sativa* L. and sainfoin *Onobrychis viciifolia* Scop., are long-lived perennial forage species of the Fabaceae family. They hold an important place in cultivated grasslands. *M. sativa* is grown in pure stands and in mixtures, typically with grasses and other legumes. In 2006, it covered 7.1 million ha in Europe and ca. 30 million ha around the world (Mouttet *et al.,* 2014). At the beginning of the 20th century, *O. viciifolia* was largely cultivated in Europe. However, over the century, the surface decreased and this decrease was exacerbated by the Common Agricultural Policy of the European Union (Frame, 2005). Although the effect of unfavourable temperature is probably more critical during germination than at any other stage of vegetative growth, the response to temperature during germination of these two pasture legumes has only been described for few varieties and a narrow range of temperatures. Modern *M. sativa* is a complex of eight diploid or auto-tetraploid perennial and allogamous subspecies native to contrasted geographical zones, in particular in terms of the annual time course of temperature. Knowing the eventual relationships between responses to temperature during germination and subsequent plant functioning could be useful for breeder. Indeed, seed germination response can be used as an early marker for selection of varieties adapted to future climatic conditions. The objective of the study presented here was to analyse germination variability of alfalfa and sainfoin in response to constant temperature.

Materials and methods

Six commercial varieties of *M. sativa* ssp *sativa* ('Barmed', 'Demnate', 'Flamande', 'Harpe' 'Luzelle' and 'Orca'), one wild population of *M. sativa* ssp *falcata* (Krasnokustkaya) and two commercial varieties of *Onobrychis viciifolia* ('Albion' and 'Canto') were studied. Seeds of *M. sativa* were obtained from Centre de Ressources Génétiques des Espèces Fourragères in Lusignan (INRA-UR4 P3F, France). Seeds of *O. viciifolia* were bought from Michel seed (Provins, France). They were conserved at 5 °C and 30% relative humidity, until they were used. Seeds of *M. sativa* were scarified between two sheets of sandpaper (grade 180) in order to break any residual seed dormancy. Further, *O. viciifolia* seeds were removed from the pod before scarification. After scarification, four sets of 100 seeds per accession were germinated in plastic Petri-dishes lids (90 mm diameter), over two sheets of Whatman paper (Whatman, France). Paper was moistened with 5 ml de-ionized water. Petri-dishes were placed in the darkness in growth chambers at the following constant temperatures: 5, 10, 15, 20, 25, 30, 35 or 40 °C and watered as needed. Vapour water deficit of the growing cabinets was kept under 1 kPa. A seed was defined as germinated when the emerged radicle was at least 2 mm long. Germination counting was carried out at variable time intervals and duration that depended on temperature treatments. Here, we report data on maximum germination percentage. For each population, a third degree polynomial was adjusted by the least squares method. From this fitting, optimal temperature for germination was estimated. Sequential ANOVA pair-wise comparisons were performed between the best fit of a given population and the rough data of a second one. The probability of a calculated F value greater that a tabular F ($Pr>F$) was calculated and a comparison matrix was constructed as previously described (Ahmed, 2015).

Results and discussion

In this report, we focus on the response of accessions to temperature during germination, in terms of percentage. The novelty of this study comes from the wide range of constant temperatures evaluated (5 to 40 °C). It was observed that most of the seven accessions of *M. sativa* were little affected by low temperature (5 °C) in their maximum germination capacity (Figure 1a to g). Exceptions to this were 'Harpe' and 'Orca'. On the other hand, germination at 40 °C was low and showed high variability in most *M. sativa* accessions. 'Demnate' had the highest germination percentage at 40 °C. For each accession, the best fit to a third degree polynomial is presented in Figure 1. Optimum germination occurred between 15.2 and 21.1 °C. The curves were used to compare the responses of these accessions. Beyond the differences in response to particular temperatures, the shapes of the best fits were significantly different ($P<0.01$) between some of the seven accessions of *M. sativa* (Figure 1a to g). For example, for the natural population Krasnokutskay, and the varieties Flamande, Demnate and Luzelle the responses were significantly different. Overall, the varieties Barmed, Harpe and Orca had exchangeable curves. An unexpected result is that the two varieties Demnate and Luzelle had a very wide range of temperatures favouring maximum germination. Further, they can be fitted linear functions ($P<0.01$) in response to constant temperature from 5 to 40 °C.

On the other hand, no significant differences ($P<0.01$) were observed between the responses of the two *O. viciifolia* varieties (Figure 1h and i). Further, these two varieties were highly sensitive to the highest temperature and germination was strongly reduced at 40 °C.

Conclusions

The finding of this work suggests that *M. sativa* accessions were different in their response to temperature during germination. Further, their responses are different from those of *O. viciifolia*. Nevertheless, while the two sainfoin varieties tested display low germination percentage at 40 °C, they have higher optimum germination temperatures (20 °C for Albion and 23.5 °C for Canto) than the alfalfa accessions tested (between 11.7 and 21.1 °C for those showing a peak). This prompted us to extend the analyses of the response to temperature during the early stages (germination and heterotrophic growth) to other forage species (Ahmed, 2015).

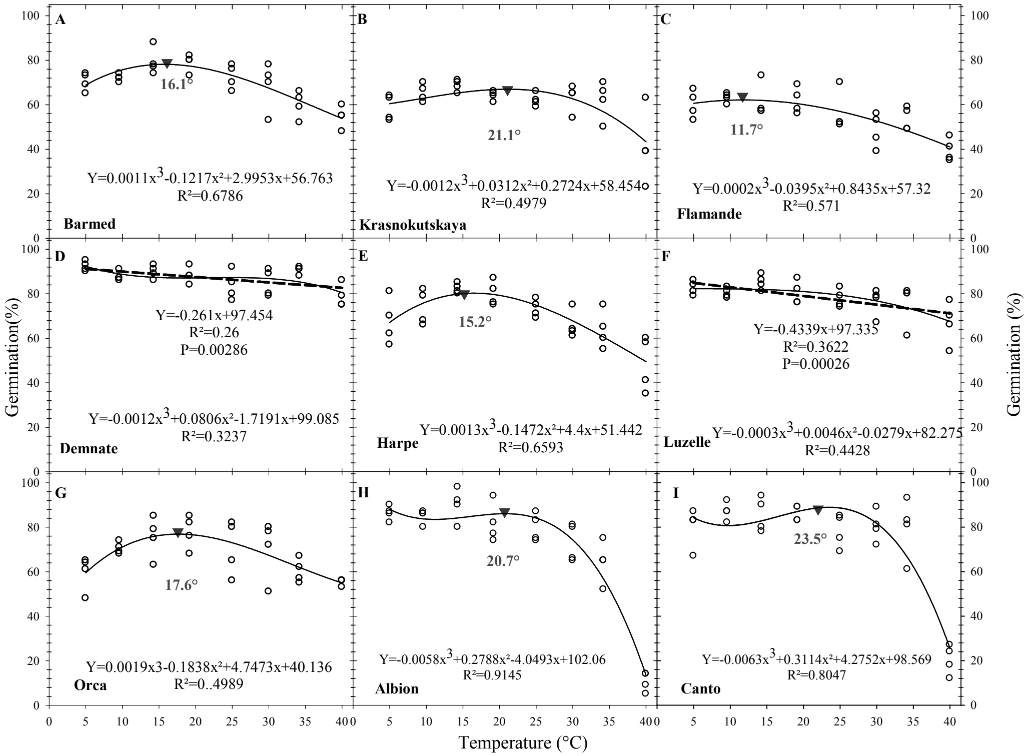

Figure 1. Germination percentage of seven accessions of *M. sativa* (A to G) and two varieties of *Onobrychi viciifolia* (H and I) in response to constant temperature. Continuous lines represent the best fit to third degree polynomials. Dashed lines, where presented, are the fitted straight line. Estimated optimal temperature is marked by ▼.

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Biofuel production from European grassland and impact on the livestock sector

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Abstract

It has been suggested that the pressure on European grassland has recently declined, while its biomass could be used for energy provision, thereby contributing to a low carbon economy. In particular, grassland feedstock can be used as a second generation biofuel, through transformation of its ligno-cellulosic content. However, it is not clear how much grassland biomass could be utilized without competing with the feeding of livestock, whose demand is expected to rise globally. Furthermore, the future of European grassland remains uncertain: while a potential increase in productivity has been suggested in both agriculturally improved pastures and extensive grassland, ambitions to halt biodiversity loss in Europe could play a role in the opposite direction. In this paper, we use the GLOBIOM global land-use model to investigate the impacts of using different amounts of grassland biomass in the EU on the grassland area and the livestock sector. Results suggest a possible but limited contribution from grassland in sustainable biofuel production.

Keywords: biofuel, grassland, Europe

Introduction

Grassland occupied 70.5 million ha within EU28 in the year 2013, i.e. 13% and 33% of total land area and total utilized agricultural area, respectively. It contributes to roughly one quarter of livestock protein needs, but recent studies highlighted a decrease in such needs, due to adjustments in the demand for meat and dairy products as well as strong shifts in the livestock sector towards forage crops and feed concentrates (Peeters, 2012). Meanwhile, climate change concerns encourage a larger use of biomass as a substitute to fossil fuels. In addition to heat and electricity, grassland-based feedstock could be processed as a 2nd generation biofuel, through transformation of its ligno-cellulose (Schubert, 2006). However, the amount of 'under-utilized' grassland that could be used for biofuel provision with limited impact on the livestock sector is not clear from available statistics. Some authors suggest the productivity of grassland could be increased (Smit *et al.*, 2008; Chang *et al.*, 2015). In addition, while the intensification of agriculture has largely contributed to the decline of European terrestrial biodiversity, European grasslands still host a large number of biodiversity rich areas (Peeters 2009). European and national policies intend to stop such trends and could seriously limit the contribution of European grassland to biofuel provision. In this paper, we explore the potential impacts of producing second generation biofuel from grassland feedstock on the livestock sector and the European grassland area.

Materials and methods

We use the GLOBIOM (Havlík *et al.*, 2014) land-use model of the agricultural, forestry and bioenergy sector to investigate the potential impacts of second generation biofuel production from European grassland biomass. In its European-zoomed version (Frank *et al.*, 2012), GLOBIOM simulates at a ten-year time step the dynamics of the demand, supply, prices and trade of agricultural goods across 57 world regions, under assumptions on exogenous drivers of the demand, trade, and the productivity and the availability of land resources. It relies upon a detailed description of the agricultural production systems at 5 arcminutes. Grassland is represented as an explicit land cover derived from Corinne Land Cover 2000 and corresponds to temporary and permanent grassland areas. The harvested productivity of grassland is derived from simulations from the EPIC and CENTURY biophysical models, with various levels of management intensity. In addition to grassland feed sources, livestock can be fed from fodder crops (corn silage, other green fodder), grains, feed concentrates and straw, with feed ration being specific to livestock species and production systems (Havlík *et al.*, 2014).

For this study, we refined the grassland productivity over Europe: at NUTS2 level, we (1) collected the EUROSTAT information on the share of rough grazing vs pasture within permanent grassland circa the year 2000; (2) used this information to generate a set of grassland productivity layers of increasing intensity (each being a weighted sum of pasture off-take – as simulated by EPIC – and rough grazing off-take – as simulated by CENTURY for natural vegetation –); and (3) selected among these layers of various intensity the one minimizing the difference to the permanent grassland productivity reported by Smit *et al.* (2008). We then simulated various scenarios to study the effect of a demand for grassland biomass dedicated to second generation biofuel production. In the 'baseline' scenario the model is run until 2030 under assumptions of the SSP2 'Middle-Of-The-Road' Shared Socio-economic Pathway (changes in demand, technological progress) and a demand for bioenergy following the Reference scenario of Capros *et al.* (2010) for Europe (with no biofuel from grassland biomass). In the 'low biofuel', 'moderate biofuel', and 'high biofuel' scenarios, we impose the production of 1.5, 3 and 6 million tonnes of fuel equivalent from European grassland. Assuming a fermentation pathway, these scenarios correspond to an inelastic additional biomass demand of respectively 8.5, 17.1 and 34.2 million tons of dry matter which has to be supplied from European grasslands. Thus, the 'moderate biofuel' scenario corresponds to both about 10% of the total target for the EU renewable fuel in transport by 2020 and about 10% of the current biomass production from European grasslands. We impose no particular distribution of this additional grassland biomass production across EU Member States and let the model simulate optimal locations.

Preliminary results and discussion

According to the model most of the new grassland biomass demand would be produced in the UK and Ireland (Figure 1A) where grassland productivity and area are known to be high, and to a lower extent in Poland, Romania, France, Germany, Spain and Sweden for 'moderate' and 'high' scenarios. While at best 25% of this biomass would come from additional grassland area, most of it would come from intensification within the livestock sector (Figure 1B), with a higher share of forage crops in the feed ration, and net fodder losses (partially compensated by concentrates and grains).

This intensification of the livestock sector leads to modest increases in prices of livestock products (up to 5.1%, Table 1) and to a small decrease in production (up to -1.7%). The total grassland area is only slightly affected (less than 1% increase) and slightly reallocated to more productive areas (about +1% in

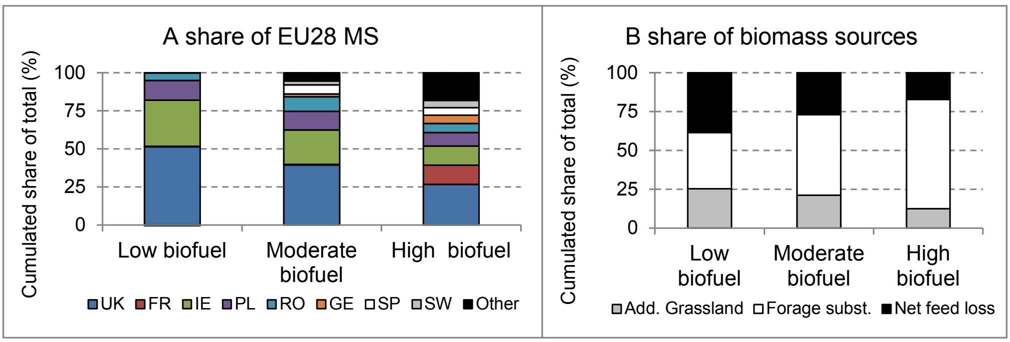

Figure 1. Decomposition of the origin of the new grassland biomass production in 2030 (A) by country and (B) by source.

Table 1. Main results: effect of scenarios on selected variables, in percentage change compared to the baseline scenario by 2030, at the EU-level.

Scenario	Grassland area	Grassland yield	Forage crop area	Fodder yield	Livestock production index	Livestock product price index
Low	+0.3%	+0.7%	+3.8%	+0.2%	-0.3%	+1%
Moderate	+0.6%	+1.1%	+15.2%	-2.9%	-0.8%	+2.5%
High	+0.8%	+1.3%	+43.2%	-7%	-1.7%	+5.1%

yield). On the other hand, the area of forage crops increases more than proportionally with the demand (up to +43%) and their yield decrease (up to -7%).

Conclusions

Model results suggest that a significant contribution of grassland to biofuel production (roughly 10% of current grassland production) is possible, however at the cost of a strong intensification of the livestock sector. Although triggering only modest impacts on the price of livestock products, this trend is more than proportional to the size of the biofuel demand, and indicates likely impacts on the environment via increased forage crop area, and potentially effects outside of Europe. We will further explore one scenario considering potential increases in grassland productivity, as it could defuse such a trade-off. Impacts on highly biodiverse grassland will also be explored.

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Climate-smart strategies to safeguard persistency and achieve sustainable grassland production

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Abstract

We report a new and innovative research project funded by the Wales National Research Network for Low Carbon Energy and the Environment and focused on improved grasslands. These dominate the Welsh landscape occupying 43% of Welsh lowlands and 26% of Welsh uplands, where they sustain livestock farming. Against a backdrop of increasing atmospheric CO_2, ground-level O_3 and temperature, the UK and many areas of Europe have recently witnessed unprecedented extreme weather patterns impacting on the capacity of agricultural grasslands to deliver a range of ecosystem services (e.g. food security, carbon and water storage, biodiversity). These atypical weather events include extreme floods, high levels of ground-level O_3, wildfires, prolonged periods of heat stress and drought. To future-proof the ecological, socioeconomic and cultural aspects of agricultural landscapes, land users will need to adapt management practices to meet this challenge. Central to achieving this goal is the adoption and implementation of holistic and novel climate-smart plant breeding programmes with soil- and animal-based management strategies to provide landscape resilience. Critically, we need to develop regimes which provide resistance to more than one stress (multi-stress resilience). The project aims to develop and validate new regimes specific to the protection of lowland productive grasslands.

Keywords: grasslands, *Festulolium*, multiple stresses, tipping-points

Background

The most recent IPCC report predicts major increases in global mean air temperatures of between 1.8 and 4.0 °C by 2100, bringing with it greater uncertainty in weather patterns and also an increased incidence in extreme events (IPCC, 2013). These changes in climate are predicted to have a major impact on the sustainability of agricultural ecosystems worldwide with the effects of climate warming already apparent (e.g. shifts in pests/diseases, changes in vegetation patterns (Dullinger *et al.,* 2012). With this gradual climate change come unpredictable and extreme weather events such as heat waves, extreme droughts, heavy rains, storms and their associated effects such as increased frequency of fires and floods, and damaging concentrations of tropospheric ozone and other gaseous pollutants. Extreme events cause instantaneous and sometimes irreversible impacts on natural ecosystems, crop productivity and human health as well as facilitating biological invasions (Ciais *et al.,* 2005). Worryingly from a food security perspective, the incidence of extreme weather events has become more frequent in the last 20 years (WMO, 2013). For example, in Europe, flood and drought events currently believed to have an intensity of 100-year are predicted to recur every 10-50 years by the 2070s. Recent examples of these include the extremely hot summer in Southern Europe in 2003, where temperatures exceeded 40 °C for several consecutive days, and the exceptional flooding seen in parts of the UK in 2012 and 2014 when water lay on the land for several months (English Nature, 2014). The immediate impact of flooding on crop production alone in 2014 in England was estimated at £25 million (ADAS, 2014). However, the socio-economic after-effects of these extreme weather events may also persist for many years after the event has occurred, and in some cases has led to destabilization of local communities (Lehner *et al.,* 2006).

It has been predicted that the biggest threat to agroecosystems in Europe is not the progressive climate change but the episodic (typically stochastic) extreme events which will occur as part of the alterations in global climate systems (Anyamba *et al.,* 2014). Despite this risk, our understanding of how extreme events will impact on plant and soil functioning and the downstream benefits/impacts (e.g. animal productivity, socioeconomics, water quality) remains poor (Grossiord *et al.,* 2014; Leingartner *et al.,* 2014). Further, the rates of ecosystem recovery after extreme conditions also remain poorly characterised. In particular, the identification of tipping points which demarcate the transition of one stable ecosystem functioning state to another is not known and this represents a major knowledge gap. There is also increased evidence to suggest that these extreme events do not necessarily occur independently from one another. For example, (a) periods of summer drought are often followed by periods of heavy rain and flooding, (b) periods of drought are often preceded and accompanied by high temperatures (heat stress) and elevated ground-level O3, or (c) agricultural systems can be exposed to both winter flooding and summer drought. Again, our knowledge of the combined impact of two or more different extreme stress events occurring in the same year (or in successive years) on the stability of agricultural ecosystems remains very poor (Grossiord *et al.,* 2014; Leingartner *et al.,* 2014).

Most extreme weather events are likely to lead to rapid and significant changes in grassland ecosystem services (supporting, provisioning, regulating and cultural services). In most cases, these events are likely to have negative outcomes. However, considerable trade-offs and unintended consequences may occur, compromising accurate predictions of long-term impacts. From a broad perspective, plants are generally considered to be more susceptible to water stress than soil microbial communities. This probably does not mean that soil microbial communities are not affected by water stress, but that their large diversity and high degree of functional redundancy may allow the community to maintain function even if some species are lost. Our field surveys have shown that recently established *Lolium perenne* swards were particularly susceptible to long-term flood damage and their recovery was extremely poor. In most cases, the sward needed complete replacement due to poor re-establishment and the dominance of weeds, which showed much greater resistance to flooding. In contrast to *Lolium perenne* monocultures, species-rich grass swards managed for biodiversity showed a high degree of resilience to flooding and recovered quickly with few adverse effects. However, these biodiverse meadows are of low productivity and not suitable for intensive agriculture.

A key challenge is to learn from these semi-natural systems how best to design sward mixes with functional traits that promote resistance to stress and ensure recovery to enable maintenance of crop productivity. Where possible, we need to select for sward mixtures that have multi-stress resistance (i.e. to drought, flood, high O3, heat stress, etc.). IBERS has led the world in *Festulolium* breeding and its importance is now becoming increasingly recognised as the grass hybrids are capable of combining the major attributes of ryegrass (high growth rates and yields of nutritious forage) with improved and deep root systems and resilience to stress found in fescues. Unique productive *Festulolium* species' combinations have been produced that include IBERS ryegrass varieties and fescue species indigenous to land areas ranging from northern Europe (*Festucolium pratensis*) through to Mediterranean regions (*Festucolium arundinacea* and *Festucolium glaucescens*) and North Africa (*Festucolium mairei*). In mixture combinations, they provide opportunities to safeguard the future for sustainable livestock agriculture during a time of climate change. They may also yield additional ecosystem service benefits including deep rooting which may promote greater soil biodiversity, soil C sequestration and greater nutrient use efficiency (Humphreys *et al.,* 2014a). Recent studies indicate that the stress tolerance inherent to *Festulolium* species' combinations may also enhance ruminant nutrient-use-efficiency, which should lower livestock greenhouse gas emissions and improve livestock gain (Humphreys *et al.,* 2014b).

We describe a new multi-discipline project that seeks to better understand the criteria that underpin grassland responses to extreme weather events and methods designed to validate the resilience of new purpose-built climate-smart grassland swards.

The project will address the following hypotheses:

- Multiple extreme weather events within a year make the ecosystem more pre-disposed to a non-reversible change in ecosystem functioning than single extreme events.
- Grassland recovery periods are much longer after multiple interacting extreme weather events than from single events.
- The duration of stress needed to induce a complete loss of vegetation function is much shorter than the time required to induce a loss in below-ground soil functioning.
- Soil mesofauna rather than soil microorganisms are most sensitive to extreme weather events and this contributes to a long-term decline in soil quality.
- Multiple extreme events result in more severe soil water repellence negatively affecting the infiltration characteristics and soil moisture storage in soils.
- More diverse grassland sward mixtures incorporating use of novel *Festulolium* hybrids have a higher resilience to extreme weather than conventional *Lolium* swards.

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Life-Dairyclim, European project aiming to mitigate methane emissions and carbon footprint of dairy cows

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Abstract

How can dairy farming contribute to reduce the climate change without compromising food security and farm economy? This is the question the project Life-Dairyclim wants to answer. The project gathers partners from research groups, association of advisory services to farmers and feed industry in collaboration with private farmers in three countries (Belgium, Luxembourg and Denmark). It focuses on production of feed, including utilisation of grassland and feeding of dairy cows in order to implement strategies that can contribute to a sustainable development of the dairy sector. Feeding experiments to decrease methane from dairy cows will be assessed at the University of Liège (Belgium) with cows milked by an automatic milking system. Methane production will be analysed individually by a device (Guardian®) inserted in the feeding bin as well as by mid infrared spectrum analysis of milk. The effect of concentrate composition on methane production during grazing in combination with optimization of grazing practices will be studied in collaboration with the industrial partner, Dumoulin (Belgium). The carbon footprint of produced milk will be determined using lifecycle assessment methods based on input from the experiments in combination with effect of feed production on especially carbon sequestration from different type of crop and utilization by Aarhus University (Denmark) and Convis, association of advisory services to farmers (Luxembourg). An important part of the project is dissemination based on pilot farms in all three countries documenting the impact of mitigation strategies adopted during the project.

Keywords: methane, GHG, LCA, life-cycle assessment, grasslands, carbon footprint

Introduction

Following the report of FAO (2014), agriculture is responsible for 12% of the production of greenhouse gases (GHG). Out of these, methane from enteric fermentation in cattle represents 74% of total emissions. Following several studies (FAO, 2010; Müller-Lindelauf *et al.*, 2010) intensification of farming is considered as a way to diminish methane emissions per kg milk. On the other hand, grasslands sequester carbon (Soussana *et al.*, 2010) and consequently can contribute to reduce the Carbon Footprint (CF) of dairy products. Furthermore, dependence to off-farm produced feedstuffs has an impact on the milk CF and could increase economic difficulties of the dairy sector because of the volatility of prices. It is thus necessary to quantify the impact of different strategies on GHG emissions by using a method that include all the emissions before and at the dairy farm including effects of feed production on carbon sequestration from different type of crop and utilization. In this context, the Life-Dairyclim project funded by the European Commission aims to develop feeding strategies to mitigate GHG emissions of dairy cows during winter indoor feeding with silage as well as during summer with pasture and to evaluate their impact on the CF of produced milk. Three countries (Belgium, Luxembourg and Denmark) involving partners from feed industry (Dumoulin), association of advisory services to farmers (CONVIS, Luxembourg), science (Aarhus University, Denmark and University of Liège, Belgium) and private farmers collaborate in this project.

Materials and methods

The project is focused on 3 objectives: First, feeding strategies likely to limit methane emissions will be tested during the winter (barn feeding) and summer (grazing and supplementary feeding) in the experimental farm of Sart Tilman (Belgium). During the winter period the cows will receive a totally mixed ration (TMR), mainly composed of maize and grass silages, representative of those observed in the dairy farms of the participating countries. The herd of 50 dairy cows milked by an automatic milking system (AMS) will be divided into two groups. Both groups will receive the same TMR, with daily registration of intake at herd level, but a concentrate of standard composition (AT1) will be allocated to one group while the other one will receive concentrate whose composition is likely to decrease methane emissions (AT2). AT1 and AT2 quantities will be determined on basis of days in milk and of mean milk yield of previous days on individual basis. During milking by the robot, several data will be recorded: milk yield, milk composition (estimation of fat and protein %), rumination time, weight, activity time. Furthermore, methane and CO_2 emissions of each cow will be registered by a device (Guardian®) installed in the feed bin of the robot. Individual milk samplings will be collected 3 times during each trial and methane emission will by estimated by near infra-red (NIR) spectrum analysis. The evaluation of CF of milk from each treatment will be performed by the lifecycle assessment methodology described by Mogensen *et al.* (2014). In summer time, the utilisation of grass will be optimized by using precision grazing methods. Total dry matter provided by the grass will be checked every week by measuring grass height on each paddock with the rising plate meter (Jenquip®). The grass height will then be multiplied by the grass cover (kg DM cm^{-1} grass) obtained by mowing a 10 meter long band, weighing the cut and drying it in oven for 72 h to estimate the DM content. Grass nutritional values will also be monitored by sampling the paddocks. All these data will be implemented in a file named 'Observatory of grass' to estimate the total DM/Ha of grass available on the farm. A strip will be used to divide pastures into smaller areas to offer the cows the most correct quantities to cover their needs and avoid spoilage. This methodology will enhance the productivity and is expected to lower CH_4 emissions/kg milk. This effect will be quantified in the Life-Dairyclim project using the methodology described by CONVIS (Lioy *et al.*, 2012). Effect of concentrates given at milking on methane emissions will be studied by collecting the same parameters as described previously. Data will be assessed to find the best compromise regarding resilience and sustainability. The CF will be calculated as described before, including the effect on pasture productivity.

The second goal will be to evaluate grasslands use and grazing practice based on a survey conducted among dairy farmers of the 3 participating countries and development in grassland productivity and area at national level based on information from national statistics. Monitoring the development in grasslands and the proportion of grazing dairy cattle in the involved countries will allow estimation of emissions accruing from land use and from land use change in the three participating countries.

Finally, dissemination of results will be a key-point of this project. The best feeding practices will be evaluated in 10 private pilot farms of the collaborating countries. In 4 of them methane emissions will be determined by the Guardian® while NIR-spectrum analysis will evaluate methane emissions in the other pilot farms. Productivity at herd, crop and farm level will be monitored each year with specific focus on resource efficiency, economic turnover and suitability indicators like fossil energy use and emission of GHG. Figures based on the method developed by Kristensen *et al.* (2011) will be faced with those obtained by Convis with the methodology described within the frame of the Interreg-Project Optenerges (Lioy *et al.*, 2012).

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Effect of cutting frequency on above- and belowground biomass production of soft rush (*Juncus effusus*) and compact rush (*J. conglomeratus*)

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Abstract

The infestation by soft rush (*Juncus effusus* L.) and compact rush (*J. conglomeratus* L.) has increased in coastal western Norway in recent decades, reducing forage quality and hampering milk and meat production. This field study explored the development of aboveground and belowground fractions of rush from seedlings to three-year-old plants, including the impact on regrowth capacity of one and two cuts per year. Soft rush showed significantly higher biomass and tussock area than compact rush after the first growing season. One annual cut in July effectively reduced biomass production in both species by 30-82%, whereas two yearly cuts in June and August gave only a small additional decrease in growth compared to one annual cut. Mechanical control measures such as cutting or mowing can thus effectively reduce rush vigour when performed in late summer.

Keywords: soft rush, compact rush, weed, grassland

Introduction

Soft rush (*Juncus effusus* L.) and compact rush (*J. conglomeratus* L.) are perennial weeds that have become more challenging in pastures and meadows in the west coast of Norway during the recent decades. Both species might be abundant or locally dominant in a range of humid habitats, however, the main impression is that soft rush is prevailing over compact rush in agricultural conditions. To optimize control methods, knowledge of these species' growth rhythm throughout the entire growing season and at which stage they are most vulnerable, is essential. The aim of this experiment was to examine the development of aboveground and belowground fractions of soft rush and compact rush from seedling stage to three-year-old plants under two cutting frequencies simulating different agricultural management systems in Western Norway.

Materials and methods

The experiment was conducted at Fureneset, Fjaler, Norway (61°34'N; 5°21'E, 10 m a.s.l.). Seeds of both rush species germinated in petri dishes and kept at 20 °C and 24 h light in April 2009 for about four weeks before being transplanted in plug trays. The seedlings were placed outdoors in mid-June and irrigated according to daily requirements until transplanted to field trials in mid-August 2009. Plants of both species were established in a complete randomised block design; three sections, each including five replicates (blocks) and 150 plants, to grow one-, two- and three-year-old plants. Both species were exposed to a cutting treatment during each of the three experimental years, imaging one- (grazing before and after the cut) and two-cut ley management in Western Norway. One third of the plants were kept uncut, one third were cut once (10 July) and one third were cut twice (10 June, 5 August). Cutting was performed by hand to a stubble height of ~7 cm, the normal mowing height in meadows. Each year, five replicate plants per species and cutting frequency (total 30 plants) were destructively sampled in (1) mid-March; (2) early June; (3) early August; (4) late September-early October; and (5) late November-early December. On each sampling date whole plants with their roots were dug from the field. The tussock area was measured (S = πab, where *S* is the area, *a* and *b* are ½ of the diagonal diameters of the tussock) and the shoots were cut off at the rhizomes. All plant material was dried at 60 °C for 48 h for dry matter (DM) determination in aboveground and belowground fractions. Analysis of variance for different plant fractions was performed separately for each year using the Proc Mixed procedure of SAS software, version 9.4 (SAS Institute Inc.) to determine effect of treatments on growth of aboveground and belowground fractions of both species. The model included species, cutting frequency and sampling date as fixed factors and replicate (block) as random effect.

Results and discussion

Soft rush had considerably more robust growth than compact rush for all growth parameters within the two last growing seasons but differences between species were greatest in the oldest plants (Figure 1). The three-year-old uncut soft rush produced on average 13 times more aboveground biomass than compact rush at the same plant age. The mean tussock area and belowground biomass of oldest plants were about five times greater than corresponding plants of compact rush. Within the growing season soft rush generally showed a higher growth in biomass at the end of the season than compact rush that tend to stagnate earlier in autumn (data not shown). These pronounced differences between the species may partly explain why soft rush comes to dominate in pastures and leys.

Cutting caused substantial reduction in both aboveground and belowground fractions of both species (Figure 1). In the oldest plants of soft rush, the mean aboveground biomass decreased by 82% after one annual cut and 89% when cut twice a year. In compact rush, the reduction in growth was slightly lower and shoot biomass of three-year-old plants decreased by about 75% after one cut and about 82% after two annual cuts. Mean belowground biomass of three-year-old plants after one cut decreased by about 59% in soft rush and 43% in compact rush, compared to controls. Two cuts caused a 9 and 25% greater decline in growth than one cut in soft rush and compact rush respectively. Mean tussock area of the oldest soft rush decreased by 63% when cut once a year and by only 5% more when cut twice a year. In compact rush, one annual cut caused about 45% decrease of tussock area and two cuts resulted in about 68% smaller tussock area, compared to control. The relatively low rush regrowth observed after one annual cut on 10 July, agrees well with our previous finding that the timing for the lowest capacity to regrowth of these two species is in mid-July-August (Kaczmarek-Derda *et al.*, 2014) and the lowest concentration of sucrose occurs in early August (Kaczmarek-Derda, unpublished results). Two cuts (10 June and 5 August) compared to one cut per year caused only a small additional reduction in growth perhaps due to still a high level of storage resources in underground fraction in June that can be used for plant regrowth.

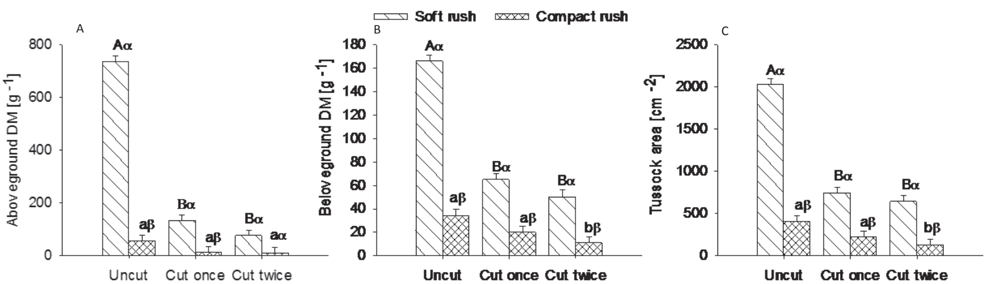

Figure 1. Aboveground biomass (A), belowground biomass (B) and tussock area (C) of three-year-old plants of soft rush and compact rush after different cutting regimes ± standard error of the mean of all sampling dates (n=25). Differences (P<0.05, Tukey test) between treatments within species are indicated by Latin alphabet (different capital letters – soft rush, different small letters – compact rush). Greek alphabet indicates differences (Tukey test) between species within treatments.

Norwegian field experiments on mowing at different times during the growing season showed that a single cut should be carried out in late summer/autumn, not in spring, and that cutting twice a year, in summer and autumn, gave the best effect (Østrem *et al.*, 2013).

Conclusions

Soft rush showed considerably more vigorous growth than compact rush within each growing season of this three-year field trial. This may partly explain why soft rush is reported as the dominant species in pastures and leys. Removal of the aboveground fraction caused substantial reductions in the belowground fractions and the reduction was higher when cutting was repeated in the following growing seasons. Both, one and two annual cuts substantially reduced the growth of the two rush species, but cutting twice at normal grass harvesting time in Western Norway (10 June, 5 August), only marginally reduced growth more than one annual cut (10 July). The additional effect of two cuts was more pronounced for soft rush than for compact rush. Thus, mechanical measures like cutting at right timing can effectively reduce rush vigour, but need to be repeated annually to achieve consistent effects.

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Red clover traits under selection in mixtures with grasses versus pure stands

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Abstract

We collected survivors of a red clover (*Trifolium pratense* L.) variety grown in plots with pure stands and clover/grass mixtures at two locations in Norway (Ås and Stjørdal). The survivors were planted in pots in the greenhouse, vernalized and allowed to flower. Plants originating from the same plot were crossed with each other using bumble bees. The offspring populations were cultivated at 16 °C and 20 h photoperiod and characterized for timing of stem elongation and above-ground biomass after 3 months. On average across replicate plots and locations, offspring of survivors from clover/grass mixtures started to elongate earlier and obtained larger above-ground biomass 3 months after sowing than offspring of survivors from pure stands. Our results suggest that intraspecific and interspecific plant-plant interactions exert divergent selective pressure on timing of stem elongation and assimilate partitioning. Hence, the relationship between these traits and persistence can differ between pure stands and clover/grass mixtures.

Keywords: assimilate partitioning, competition, earliness, phenological development, stem elongation, *Trifolium pratense*

Introduction

Better persistence is a major breeding goal, second to yield, in many red clover breeding programmes (Annichiarico *et al.,* 2015). It is a very complex trait, the underlying factors depending on the prevailing environmental conditions and managements. We are trying to identify traits associated with persistence by characterizing traits in survivor populations. We have collected individuals from survivor populations in field plots, generated a new generation for phenotypic characterization in order to remove effects of phenotypic plasticity, and are currently characterizing the offspring populations for various traits. Here we report on earliness of elongation and shoot biomass accumulation.

Materials and methods

Red clover (*Trifolium pratense* L.) survivors were collected from experimental plots that had been established in 2010 at two locations: Ås (59°40'N, 10°47'E) and Stjørdal (63°30'N, 10°51'E). The plots were either pure stands or species mixtures, sown at a rate of 20 (Ås), 15 (pure stands at Stjørdal) or 24 (mixtures at Stjørdal) kg ha^{-1}. At Ås the mixtures consisted of perennial ryegrass, tall fescue, white clover and red clover (each species constituted 25% of the total seed weight sown). At Stjørdal the mixtures consisted of timothy (54%), meadow fescue (25%) and red clover (21%). The plots were harvested three times per year from 2011 to 2013, and fertilized with a compound fertilizer at a rate of 100 (Ås) and 130 (mixtures, Stjørdal) kg N ha^{-1} year^{-1}. Pure stands at Stjørdal were applied a fertilizer without N. In October 2013 sixty survivors were collected from each of two replicated plots in Ås while forty survivors were collected from each of three replicate plots at Stjørdal. The survivors were planted in large pots and kept in a greenhouse at Ås over the winter at a cool temperature (minimum 2 °C). Prior to flowering the following year survivors from each plot, considered a population here, were covered with a large net and one bumble bee nest with 50 bees (Pollinering AS, Bryne, Norway) was placed inside each net. After maturation seeds were collected from each population. In the beginning of 2015 two week old seedlings were grown under controlled conditions at 16 °C and 20 hour photoperiod at 240 μmol m^{-2}

s^{-1} photosynthetically active radiation (PAR) and a red light to far red light ratio (R:FR) of 2.25. After 1 month half of the plants were moved to a shade treatment with 100 µmol m^{-2} s^{-1} PAR and a R:FR of 1.15 (16 plants per population and light treatment). The number of days from planting of seedlings until stem elongation (DTE) was counted. Plants that did not elongate were given the maximum value + 1 (76). Three months after sowing the plants were cut at 5 cm height and the dry weight (DW) was recorded. Analysis of variance and regression analysis were performed in SAS 9.4 (SAS Institute Inc., Cary, NC, USA) with average values for each population and light treatment, using PROC GLM and PROC GENMOD. The model for regression analysis was DW = DTE + light treatment (LT) + location (L) + stand type (ST) + DTE × LT + DTE × L + DTE × ST. Non-significant parameters were removed from the model in a stepwise manner until only significant parameters ($P<0.05$) remained.

Results and discussion

There were no significant effects of light treatment on DTE. On average, survivor populations from mixed stands started to elongate 3.8 days earlier ($P<0.05$) than populations from pure stands, but there was no significant effect of location (Figure 1A). The average DW in the shade treatment was 3.8 g plant^{-1} lower than in the light treatment ($P<0.0001$). Survivor populations from mixed stands had 1.4 g plant^{-1} higher DW ($P<0.004$) than populations from pure stands, while populations from Stjørdal had 1 g plant^{-1} higher DW than populations from Ås ($P<0.04$) (Figure 1B). There were no interactions between factors. DW decreased with increasing DTE, and more so in light than in shade (Figure 1C). When accounting for the effect of DTE on DW, there was still a significant effect of both location and stand type, with populations from mixed stands having 0.9 g plant^{-1} higher DW than populations from pure stands, and populations from Stjørdal also having 0.9 g plant^{-1} higher DW than populations from Ås (Table 1). Our results suggest that early elongation is favourable for survival of red clover in mixtures with grasses, and/or that later elongation is favourable in pure stands. In addition, early accumulation of shoot biomass, irrespective of earliness, appears to be more favourable in mixed than in pure stands. Timothy and perennial ryegrass establish faster than red clover, and also elongate earlier in the season. Thus, competition for light may have exerted a selective pressure for early biomass accumulation and elongation in mixed stands.

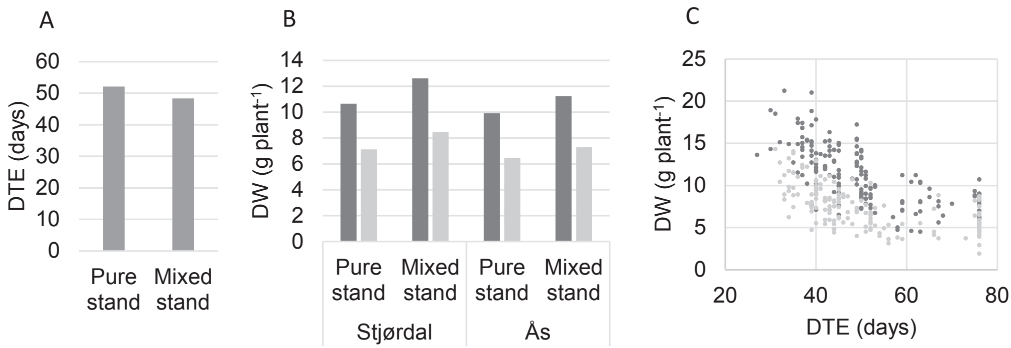

Figure 1. (A) Days to elongation (DTE) and (B) shoot dry matter above 5 cm (DW) of red clover survivor populations originating from pure stands and mixtures with grasses at two locations. (C) Interaction between DTE and DW. Plants were grown at 16 °C and 20 h photoperiod in light (black) or shade (light grey). For DTE there was only a significant effect of stand type, so the averages of location and light treatments are shown.

Table 1. Regression analysis of dry weight of shoots above 5 cm height (DW) in red clover survivor populations originating from two locations (L, Ås and Stjørdal) and two different stand types (ST, pure stands and mixtures with grasses). The plants were grown in two different light treatments (LT, light and shade).

Parameter	d.f.	Estimate	*P*-value
Intercept	1	10.797	<0.0001
DET	1	-0.086	0.08
LT (light vs shade)	1	10.558	0.001
L (Stjørdal vs Ås)	1	0.930	0.0007
ST (mixture vs pure stand)	1	0.854	0.003
DTE × LT (light vs shade)	1	-0.138	0.03

Conclusions

While red clover is almost entirely cultivated in mixtures with grasses, breeding and variety testing of red clover is almost entirely performed using pure stands. Our results suggest that this practice may not identify the genetic material with optimal persistence in mixed stands, and that there may be selection occurring in seed production fields that has a negative effect on persistence in mixed stands.

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Impact of waterlogging under different temperatures on hardening and freezing tolerance of timothy (*Phleum pratense*)

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Abstract

Precipitation has generally increased in Norway during the last decades, and climate projection indicate a further increase in the future. The growing season has also become longer with higher temperatures, particularly in autumn. Previous studies have shown negative effects of higher temperatures and a temperature-dependent effect of waterlogging on hardening capacity of timothy. We studied effects of waterlogging on seedlings of timothy (*Phleum pratense*, cv. Noreng) under three temperatures: 3 °C, 7 °C, 12 °C, and natural light in autumn in a phytotron at Holt, Tromsø (69°N). After temperature treatments, all plants were further acclimated at 2 °C for three weeks. Freezing tolerance was determined as LT_{50} and regrowth after seedlings were frozen at several predefined freezing temperatures in a programmable freezer. After freezing, survival and regrowth of new leaves was measured after three weeks at 18 °C, 24 h light in a greenhouse. Rising temperatures had a clear negative effect on freezing tolerance. Waterlogging had no effect on frost tolerance of plants grown under constant 3 °C, but had clear negative effects under 7 °C, and even clearer effects under 12 °C compared to plants not waterlogged. The results indicate that waterlogging under higher autumn temperatures may have negative implications for hardening of timothy.

Keywords: flooding, winter acclimation, climate change, autumn temperatures

Introduction

Temperatures have increased during the last decades, and spring and autumn has become warmer in northern latitudes (Bauer-Hanssen *et al.*, 2015). Precipitation has also increased in the same regions, more in autumn and spring than in winter and summer (Bauer-Hanssen *et al.*, 2015). There are more heavy precipitation events, which increases the risk of flooding (Hartman *et al.,* 2013). Dalmannsdottir *et al.* (2015) showed that increasing autumn temperatures prior to winter acclimation led to poorer hardening and frost tolerance in timothy (*Phleum pratense*). In another study, the effect of waterlogged soils during autumn on hardening seemed to depend on temperature (Dalmannsdottir *et al.,* 2012). Due to slow diffusion of oxygen in water, the exchange of air with the atmosphere is strongly reduced in flooded soils. This can lead to hypoxic or anoxic conditions, especially under higher temperatures when metabolic activity of plants is increased. In this study, we examined the effect of waterlogged conditions under differing temperatures on hardening determined as frost tolerance of timothy.

Materials and methods

Timothy seedlings (cv. Noreng), sown July 13, 2015 in a greenhouse, were transplanted into 12 cm pots in mid-August. August 25 the seedlings were transferred to a phytotron at Holt, Tromsø (69ºN) at 12 °C, under ambient light conditions. September 11 the seedlings were subjected to three different pre-acclimation temperatures, 3 °C, 7 °C and 12 °C (ambient light) for 30 days. At the same time, all plants were waterlogged by placing pots in trays, filled with water, generating completely waterlogged conditions for the roots. As control, we placed the same number of pots in trays, and watered only when required. After the temperature treatments, all plants were acclimated for 3 weeks at 2 °C under ambient light conditions. Plants were then cut 3.5 cm above soil and placed in programmable freezers and frozen, four pots per temperature and treatment, to pre-determined temperatures ranging from -11 to -26 °C. The cooling rate was -1 °C h^{-1} until it reached -10 °C; thereafter the rate was -3 °C h^{-1}. A subset of plants from each treatment was placed at 2 °C and darkness for comparison. After freezing, plants were thawed at 2 °C and darkness, and then transferred to a greenhouse (18 °C, 24 h light) for three weeks. We recorded survival of plants and calculated the LT_{50} by probit analyses using the logistic distribution in PROC Probit (SAS, 2012). Fiducial limits (α=0.05) were used to test any significant differences between treatments. Regrowth of surviving plants was measured after cutting at 3.5 cm and measuring new biomass (DM, dry matter). Significant differences between treatments were determined with ANOVA using the GLM procedure of SAS (2012).

Results and discussion

Warm temperatures before winter acclimation had a clear negative effect on freezing tolerance, and this is in agreement with results of Dalmannsdottir *et al.* (2015) (Table 1, Figure 1). Waterlogging reduced freezing tolerance measured as LT_{50} to some extent, but the effect was not significant.

Regrowth capacity after the freezing treatment was clearly affected by both pre-acclimation temperatures and waterlogging (Figure 1). Increasing pre-acclimation temperatures generally led to poorer regrowth after freezing (P<0.001). Plants at pre-acclimation temperature 12 °C had significantly higher shoot DM than at 7 °C before freezing tests (1.39 vs 1.06 g pot^{-1}). Thus, the plants at the higher pre-acclimation temperatures may have allocated more carbohydrates from photosynthesis to growth in contrast to plants at lower temperatures that may have allocated more to storage. Waterlogging affected regrowth in plants

Table 1. Tolerance to freezing (°C, LT_{50}) of timothy plants (4 pots with 5 plants per freezing temperature, 150 plants in total per treatment) waterlogged or control (watered when required) and treated with three pre-acclimation temperatures before winter acclimation.

Pre-acclimation Temperature °C	Control			Waterlogged		
	LT_{50}	Fiducial limits		LT_{50}	Fiducial limits	
3	-27.08	-25.3	-32.4	-25.40	-24.0	-28.0
7	-26.12 [1]	NS		-26.12	-24.3	-30.2
12	-21.45	-20.2	-23.2	-19.49	-18.4	-20.7

[1] The LT_{50} was not significant (NS) due to poor curve fit in the probit analysis.

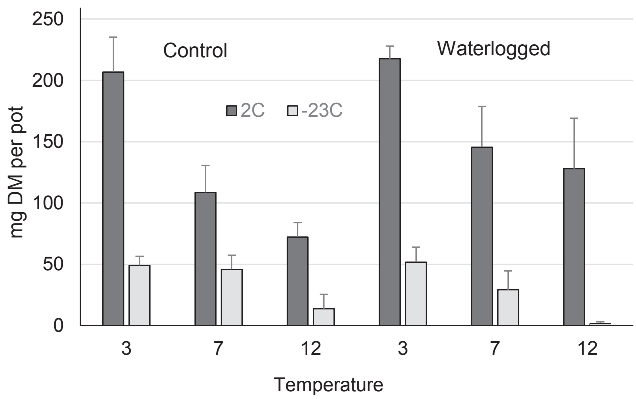

Figure 1. Leaf regrowth (mg dry matter (DM) per pot) of plants frozen at -23 °C or left at 2 °C for 48 h of plants waterlogged or watered as required (control) at three different pre-acclimation temperatures; 3, 7 or 12 °C. The regrowth is recorded after three weeks at 18 °C (n=4 pots with 5 plants per treatment, error bars are standard errors).

subjected to freezing tests or not frozen (2 °C) differently (*P*<0.04) (Figure 1, data shown only for 2 °C and -23 °C, for brevity). Waterlogged plants had higher regrowth in non-frozen plants than control plants. In plants subjected to freezing, the effect was opposite, and more negative for plants grown under 12 °C than under 7 or 3 °C pre-acclimation temperature.

Conclusions

The results indicate that the effect of waterlogging on hardening of timothy is dependent on pre-acclimation temperatures. Under higher autumn temperatures, waterlogging may have negative implications for the hardening of timothy.

Acknowledgements

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Satellite data for monitoring of European grasslands – new tool for adaptation to climate change

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Abstract

Grasslands are significant as a source of forage for animal production, but are also important in many ecological functions. To be able to analyse changes in environmental conditions of grasslands, monitoring of grassland areas using remote sensing is an important task. Studying changes in environmental condition over time and space in grasslands has been the subject of research at different scales. Such an example is the Polish-Norwegian Research Project FINEGRASS „Effect of climatic changes on grassland growth, its water conditions and biomass'. *In situ* measured soil-vegetation parameters and satellite observations have been combined and analysed to quantify the spatial and temporal variability of grassland conditions, as reflected in variations of vegetation surface temperature, soil moisture, and biomass. Results show a significant trend of increasing grassland surface temperature in Poland, based on AVHRR satellite data; a positive significant relationship between the (April-September) standardized precipitation evapotranspiration index (SPEI) and grass yields in Poland; northern Norway has shown trends towards warmer springs and autumns since 1991, and significant trends towards earlier snowmelt and green-up on test fields in northern Norway.

Keywords: grassland, remote sensing, surface temperature, soil moisture, biomass

Introduction

During recent years, European grassland area has been reduced as a result of intensification of grassland and animal production, abandonment or areas turned into bioenergy crops, and effects of European Union policy (Huyghe *et al.*, 2014). Permanent grasslands cover 33% and temporary grasslands cover 6% of the total agricultural area in Europe. To analyse changes in grassland area and productivity, a constant monitoring of grasslands is important. Remote sensing provides the opportunity to monitor grassland environmental conditions spatially with high revisit frequency. The European Copernicus Programme gives possibilities for grassland monitoring, including the application of new satellite data from the group of Sentinel satellites. The Polish-Norwegian Research Project FINEGRASS „Effect of climatic changes on grassland growth, its water conditions and biomass' has focused on detecting grassland development and change in Poland and Norway by applying different approaches based on site-specific environmental and agricultural conditions. The prime objective of the FINEGRASS project is to develop remote sensing techniques as tools for assessing the influence of climatic changes on grassland growth, water conditions and biomass production. This paper addresses changes in grassland surface temperatures in Poland, the relationship between standardized precipitation evapotranspiration index (SPEI) and grass yields in Poland, and phenological trends of grasslands in northern Norway.

Materials and methods

For the Poland study sites, a grassland layer was derived from CORINE Land Cover (CLC) data. The surface temperature (Ts) for each grassland pixel, collected from AVHRR (since 1997) satellite data, was overlaid on the CLC grassland map. The surface temperature was taken from the 10-day composite to correct for atmospheric influence on temperature values. For each of the administrative divisions (NUTS1), the surface temperature was averaged for the grassland pixels, and the trend in surface temperature between 1997 and 2015 was analysed in each NUTS1 region.

Annual dry matter yield data were collected from ca. 50 ha of grassland situated on mineral soils, utilized mainly for grazing, and of ca. 150 ha of grassland located on organic soils used exclusively for cutting for feed. The effect of climate conditions over a 50-year period on yield of grasslands located on mineral and organic soils in central Poland were analysed. Plot-level yields were compared with long-term averages of mean monthly and annual temperatures, monthly precipitation sum and standardized precipitation evapotranspiration index (SPEI), which incorporates both these climatic elements and characterizes drought severity. The SPEI uses a difference between precipitation, expressed as the standardized precipitation index SPI, and potential evapotranspiration (PET).

Meteorological data were used from the Holt research station in northern Norway to assess temperature trends in spring, summer, and autumn since 1991. MODIS 16-day NDVI data were also used in conjunction with a ground-truth phenology dataset at Holt to assess trends in snowmelt and green-up between the years 2000 and 2012.

Results and discussion

Figure 1 presents the trend in surface temperature of the studied grasslands from the years 1997-2015, derived from the AVHRR data for Poland. Figure 1 shows that there is a significant increasing trend in surface temperature in the 11th 10-day period of the growing season. Weather conditions, characterized by SPEI, had a significant effect on grassland yield for grasslands located on mineral soils in all analysed periods, particularly for the period April-September (Figure 2).

The Holt temperature data showed a trend towards earlier growth start in spring and increasing temperature sum in the growing season during the last 10 years, however, with large variation between years. Temperature of summer months (June and July) do not seem to be higher than during the reference period 1961-1990, but spring months (April and May) and autumn months (August and September) have a clear increasing trend. Preliminary analyses using the MODIS 16-day NDVI time series show consistent trends at the Holt research station in northern Norway towards earlier snowmelt and green-up in the early 2000's, somewhat later snowmelt and green-up in the late 2000's, and then again towards earlier snowmelt and green-up in the early 2010's (Figure 3).

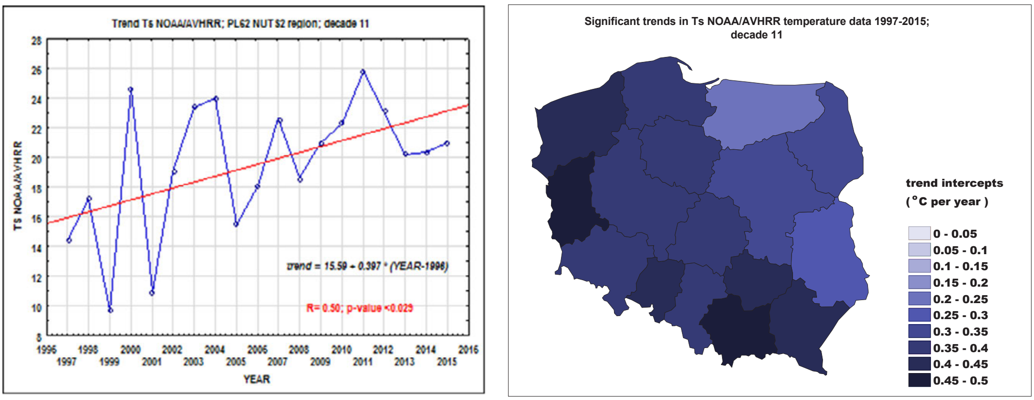


Figure 1. Significant trend of grassland surface temperature (Ts) in Poland based on NOAA/AVHRR.

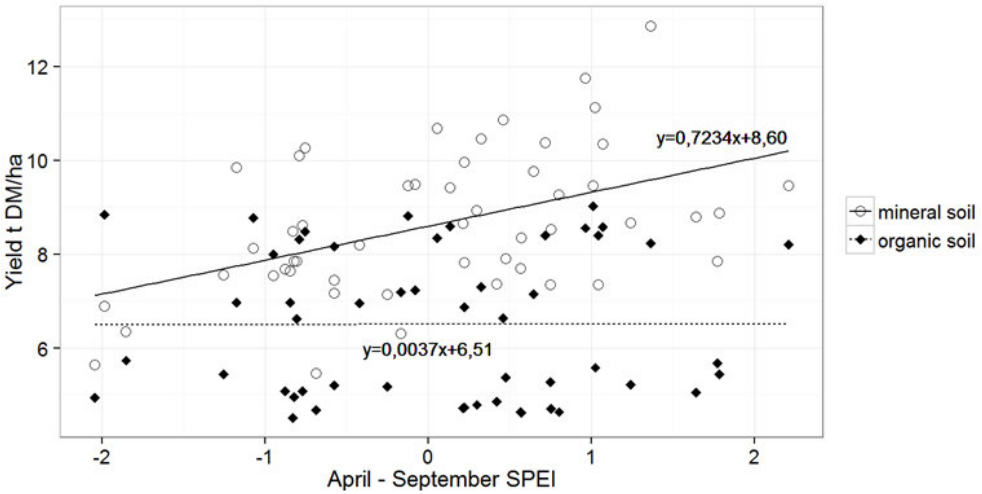
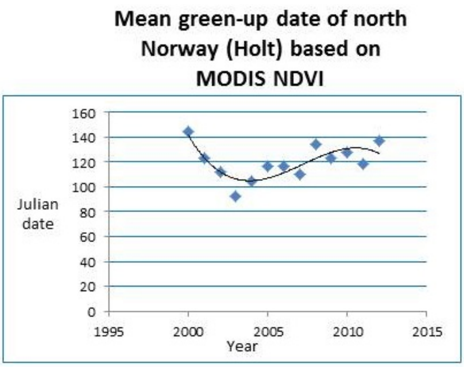


Figure 2. Relationship between annual grassland productivity and SPEI for the 6-month period from April to September, stratified by type of soil. Dm = dry matter.



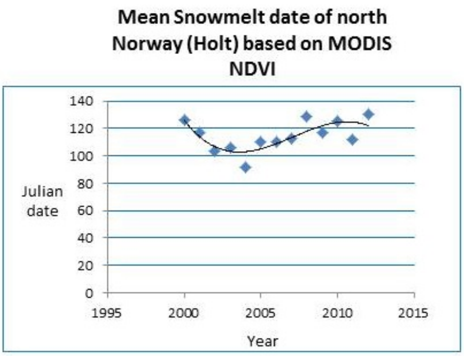


Figure 3. Trends in green-up and snowmelt (Julian) dates in north Norway based on MODIS NDVI, using a historical in-situ phenology dataset as ground truth.

Conclusions

With the recent launch of Sentinel satellites providing increased temporal resolution of landscape-scale satellite data, and with increased spectral resolution, more relevant information will be provided for monitoring effects of climate change on biomass, water cycle, and exchange of energy between grassland surfaces and the atmosphere. The preliminary results for Poland show a significant positive interaction between precipitation and yields on mineral soils, while in northern Norway there is a significant trend of increase of temperature in early spring and earlier growth start as well as increasing temperatures in autumn months.

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The effects of legume content and drought on symbiotic N_2 fixation and herbage nitrogen yield

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Abstract

Increased incidence of drought, as predicted under climate change, highlights the need to design grassland management systems for forage production that are adapted to future climate scenarios. Compared to monocultures, simple grassland mixtures can result in increased yields, greater stability in response to disturbance (i.e. drought), reduced invasion by weeds and improved nutrient retention. In particular, the interaction between species with and without the ability for symbiotic N_2 fixation (SNF) increases yield benefits. This study assessed the effect of summer drought on the N yield of monocultures and mixtures with experimentally varied legume proportions. At Tänikon research station in 2011, we used rain-out shelters to impose a 10-week summer drought on grassland monocultures and mixtures consisting of two legumes (*Trifolium repens* L. and *Trifolium pratense* L.) and two non-legumes (*Lolium perenne* L. and *Cichorium intybus* L.). We used measurements of SNF (^{15}N isotope dilution method) to reveal how legumes and non-legumes adjust their N nutrition in response to drought and legume proportion. The combination of legumes and non-legumes in simple grassland mixtures had a positive effect on N yield under both control and drought conditions. Measurement of SNF provided valuable insights into the processes behind the observed resistance to drought.

Keywords: *Trifolium repens, Trifolium pratense, Lolium perenne, Cichorium intybus,* symbiotic N_2 fixation, drought

Introduction

Climate models predict an increase in both the frequency and the intensity of extreme weather events such as droughts. This highlights the need to design grassland management systems for forage production that are adapted to future climate scenarios. Simple grassland mixtures in comparison to monocultures can result in increased yields, greater stability in response to disturbance (i.e. drought), reduced invasion by weeds and improved nutrient retention (Finn *et al.*, 2013; Hooper *et al.*, 2005). In particular, the interaction between species with and without the ability for symbiotic N_2 fixation (SNF) increases yield benefits (Nyfeler *et al.*, 2011). This study assessed the effect of summer drought on the N yield of single-species and mixed-species communities with experimentally varied legume proportions. We used ^{15}N isotope dilution method to measure SNF in order to reveal how legumes and non-legumes adjust their N nutrition in response to drought and community legume proportion.

Materials and methods

In August 2010, monocultures and mixtures were sown in 66 plots at Tänikon research station, Switzerland in a completely randomised design with 3 replicates (Hoekstra *et al.*, 2015). Four species were selected based on their ability for symbiotic N_2 fixation, two legumes (*Trifolium repens* L. and *Trifolium pratense* L.) and two non-legumes (*Lolium perenne* L. and *Cichorium intybus* L.). Plots were sown to consist of four monocultures, six binary communities (50% of two species, based on monoculture seed rates), and one equi-proportional community (25% of each of four species). Half of the plots were subjected to a drought treatment of 10 weeks of rain exclusion in 2011, using rainout shelters. Plots were harvested five times and received 145 kg N ha^{-1} yr^{-1} split over five applications.

We used the isotope dilution method to quantify SNF: A solution of double ^{15}N labelled (98 Atom%) ammonium nitrate was injected at 5 cm depth in a 50×50 cm sub plot within each 3×5 m plot. At the end of the drought treatment, the sub-plots were harvested at 6 cm height and separated into component species, and samples were dried and subjected to ^{15}N and total N analysis. The remainder of the plot was harvested with a plot harvester and a subsample taken for DM and total N analysis. We used the following model (based on Nyfeler *et al.*, 2011) to assess the effect of drought and legume proportion (Legume):

$$y = \alpha + \beta_1 \cdot drought + \beta_2 \cdot legume + \beta_3 \cdot legume^2 + \beta_4 \cdot legume \cdot drought + \beta_5 \cdot legume^2 \cdot drought$$

Results and discussion

There was a significant ($P<0.001$) effect of drought on total N yield, which was reduced by on average 17 kg N ha^{-1} cut^{-1} under drought conditions (Figure 1A). The reduction was highest in non-legume stands (61%) and lowest for the mixed and pure legume stands (23 and 18%, respectively). The N yield from SNF was not affected by drought, which could be attributed to the higher percentage of legume N derived from SNF (%SNF) under drought compared to control conditions (Figure 1D).

Total N yield of the sward increased with increasing sown legume proportion ($P<0.001$), and the ranged from 19.5 kg N ha^{-1} per cut for swards with no legumes to 82 kg N ha^{-1} per cut for swards with only legumes (Figure 1A). The N yield in mixed swards (legume proportion 0.5) was higher than expected from the proportional contributions of the non-legumes and legumes, indicating that positive mixing effects (overyielding) were occurring, as evident from the highly significant non-linear legume effect (Figure 1A). For mixed swards containing 50% legumes, the overyielding was on average 26%. The overyielding could be mainly attributed to the higher than expected N uptake by non-legumes in mixed swards (Figure 1B), indicated by a highly significant ($P<0.001$) non-linear relationship between legume proportion in the sward and N yield of non-legumes. This may be the result of the competitive advantage of non-legumes to take up N, which can lead to 'nitrate sparing' (Nyfeler *et al.*, 2011). At the same time, legumes maintained their N yield (Figure 1C) by increasing the %SNF (Figure 1D). This fits well with other evidence that legumes regulate their activity of SNF to close the gap between their N-demand for growth and the availability of N from non-symbiotic sources. In conditions with low availability of soil

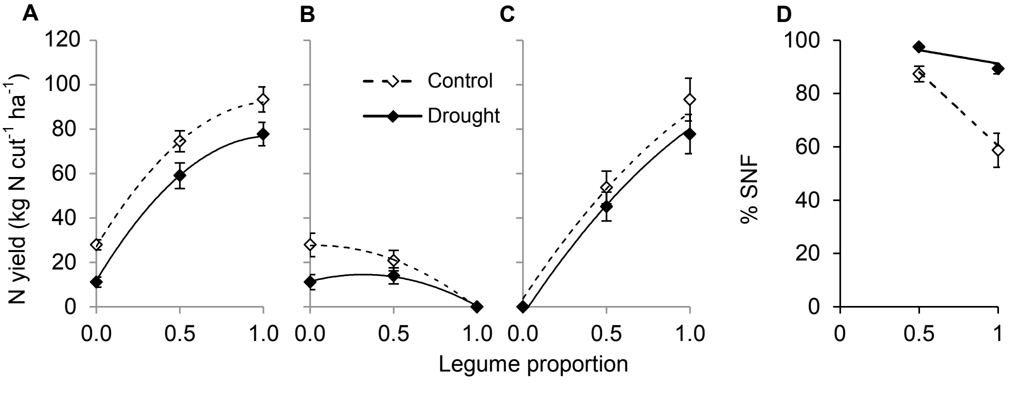

Figure 1. (A) Total N yield (kg N cut^{-1} ha^{-1}), (B) non-legume N yield, (C) legume N yield and (D) % legume N derived from symbiotic N_2 fixation (SNF) as affected by the legume proportion under control and drought conditions during the second harvest of the drought period. Data points represent the treatment mean (± standard error).

N (i.e. drought conditions and swards with a high proportion of non-legumes), legumes were mainly depending on SNF, whereas in conditions with higher soil N availability (i.e. non-drought conditions and swards with a high proportion of legumes) the %SNF was down to 60% (Figure 1D).

Conclusions

The combination of legumes and non-legumes in simple grassland mixtures maintained their positive effect on N yield under drought conditions. Measurement of SNF provided valuable insights into the processes behind the observed responses to drought.

Acknowledgments

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Climatic adaptation of species and varieties of grass and clover in the West Nordic countries

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Abstract

Eight experiments with cultivars (cvs.) of grass and clover species were established in the Faroe Islands, Greenland, Iceland, Norway and Sweden to evaluate important forage species and cvs. in terms of yield potential, persistence and adaptation to variable climate in the West Nordic countries. Timothy had on average the highest spring cover after three years of trial together with smooth meadow grass (cv. Knut), whereas perennial ryegrass had the lowest spring cover after three years. On average cocksfoot (cv. Laban) and timothy (Grindstad related cvs.) gave the highest yield, 8.85 and 8.71 t ha^{-1}, respectively, and smooth meadow grass and common bent grass the lower yields, 7.52 and 7.30 t ha^{-1}, respectively. The results from these experiments show that we have a wide range of species and cultivars usable in the West Nordic areas. We can meet an increase in temperature to a certain level by moving the more southern species and cvs. farther north, however, our most winter hardy cvs. are still important to maintain.

Keywords: climate change, winter tolerance, yield potential

Introduction

Climatic conditions, characterized by moderate to low summer temperature and very variable winter conditions, limit the potential of crop production in the West Nordic countries. Under these climatic conditions, husbandry based on grass and clover forage is of greatest importance. As the plants have to survive harsh winter conditions the grass and clover species used must have great tolerance to frost, ice cover and temperature fluctuation (Gudleifsson *et al.,* 1986) as well as diseases (Larsen, 1994). Furthermore, the grasses have to tolerate intensive grassland management (Hermannsson and Helgadóttir, 1991). It is therefore important to consider both climate and management when evaluating forage species and varieties for use in these countries. There is a general consensus among scientists that the temperature on earth is increasing because of the green house effects. Small temperature changes can strongly affect agricultural production and may affect the choice of species and cultivars. If climate conditions within the West Nordic area change as a result of global warming it is important to get information on potentials and limitations of species and cultivars concerning growth under varied climatic conditions. The main goal of this project was to evaluate important forage species and cultivars used in the West Nordic countries for future use and to find which cultivars have a wide adaptation and can be used in a wide range of climatic conditions as well as elucidating limitations to specific conditions.

Materials and methods

Field experiments were established at seven locations in the West Nordic countries and in Sweden in 2009 and 2010 (Table 1). The experimental design consisted of randomized blocks with three replicates and plot size around 10 m^2. Winter survival was observed for three years. The experiments at Korpa and Fureneset were harvested for three years and two years at the other locations. At Fureneset and Lännäs

Table 1. Locations of the field experiments.

Location	Latitude	Longitude	Altitude, m
Korpa, South-west Iceland	N 64° 09′	V 021° 45′	45
Möðruvellir, Northern Iceland	N 64° 34′	V 021° 45′	12
Upernaviarsuk, Greenland	N 60° 44′	V 045° 53′	15
Qassiarsuk, Greenland	N 61° 09′	V 045° 30′	20
Kollafjørður, the Faroe Islands	N 62° 06′	V 006° 58′	15
Holt, Tromsø, Northern Norway	N 69° 39′	E 018° 55′	15
Fureneset, Western Norway	N 61° 34′	E 005° 21′	10
Lännäs, Northern Sweden	N 63° 09′	E 017° 39′	31

the trials were harvested 3 times each year and 2 times at the other locations. Yield was not measured in Greenland because of damage that occurred in the experiments.

Twenty four cultivars (cvs.) of grass and clover species were included (country origin in brackets): Timothy (*Phleum pratense*) Snorri (Nordic), Noreng (NO), Grindstad (NO), Lidar (NO), Rakel (SE), Switch (SE); smooth meadow grass (*Poa pratensis*) Kupol (SE), Knut (NO); meadow fescue (*Festuca pratensis*) Norild (NO), Kasper (SE); tall fescue (*Festuca arundinacea*) Swaj (SE); *Festulolium* Felina (DK); cocksfoot (*Dactylis glomerata*) Laban (NO); perennial ryegrass (*Lolium perenne*) Birger (SE), Ivar (NO), Figgjo (NO); common bent grass (*Agrostis capillaris*) Leikvin (NO); red clover (*Trifolium pratense*) Torun (SE), Yngve (SE), Lavine (NO), Lea (NO); white clover (*Trifolium repens*) Norstar (NO), Litago (NO); alsike clover (*Trifolium hybridum*) Alpo (NO). Red clover and alsike clover were sown in a mixture with timothy (cv. Korpa (IS)) and white clover in a mixture with smooth meadow grass (cv. Sobra (SE)). Additionally, a mixture of four species (25% of each) was included consisting of timothy (cv. Korpa) + smooth meadow grass (cv. Sobra) + white clover (cv. Norstar) + red clover (cv. Lea).

Results and discussion

Dry matter yield (DMY) was on average highest in cocksfoot and timothy (Grindstad related cvs.), 8.85 and 8.71 t ha^{-1}, respectively, followed by tall fescue, *Festulolium*, perennial ryegrass, northerly timothy and meadow fescue, yielding 8.51, 8.47, 8.23, 8.18 and 7.98 t ha^{-1}, respectively, whereas smooth meadow grass and common bent grass gave lower DMY, 7.52 and 7.30 t ha^{-1}, respectively. The lowest yielding grass species gave 1.5 t ha^{-1} lower DMY than the highest yielding ones. The red clover cvs. yielded 6.6 t ha^{-1} and the other clover cvs. 5.4 t ha^{-1}, so red clover in a mixture with timothy gave 2.0 t ha^{-1} less DMY than pure timothy.

Spring cover after three winters was on average highest in timothy cvs. together with smooth meadow grass (cv. Knut), in which timothy (cv. Snorri) showed highest spring cover followed by the cvs. Knut (smooth meadow grass), Noreng (timothy), Laban (cocksfoot), Kasper (meadow fescue), Kupol (smooth meadow grass) and Leikvin (common bent grass). The cultivars of perennial ryegrass had generally low spring cover, however, to varying degree. The clover cvs. were not comparable to the grasses because they were sown in a mixture with grasses. Most of the clover cvs. had around 30% spring cover, with alsike clover (cv. Alpo) lowest at 21%.

The widest adaptation was seen in timothy which survived relatively well at all locations, however, with some variation between the cultivars. In general timothy was among the highest yielding species but not the highest at all locations. If timothy cvs. survive at different locations, they tend to give the same DMY ranking at all locations. Even though timothy is winter hardy it does not live for many years with early cutting dates as is practiced in some West Nordic countries, and breeding of timothy that will tolerate more cuttings and with increased regrowth capacity is indeed important to fit in an extending growing season.

Smooth meadow grass also demonstrated a wide adaptation but compared to timothy it seems to be more important to use the right cultivar at different locations. Smooth meadow grass was generally lower yielding than other species in the experiments and will probably mainly be needed in a cold climate and where grass fields are grazed. The cocksfoot (cv. Laban) was surprisingly stable over locations in this study even though it is not as winter hardy as timothy and among the highest yielding cultivars. The feeding value in cocksfoot depends, however, on early cutting to provide acceptable values. Cocksfoot might need more attention at certain locations in the Nordic countries.

Meadow fescue seemed to be more unstable than timothy and should be avoided where there is a risk of long standing ice cover. When grown under preferable conditions meadow fescue is high yielding with good feeding value. Nordic bent grass used to be winter hardy but like some other species it responds selectively to climate, being more a maritime species than continental, and due to a moderate DMY potential and feeding value it will not be widely used. Perennial ryegrass, tall fescue and *Festulolium* were not among the most winter hardy species in these experiments. However, they were high yielding at locations where they survived. Perennial ryegrass has in addition a good feeding value. Detailed results from these experiments have earlier been presented (Thorvaldsson *et al.,* 2015)

Conclusions

A wide range of species and cultivars are usable for growing in the West Nordic areas. We can meet an increase in temperature to a certain level by moving the more southern species and cultivars farther north but this can be limited because of factors such as day length requirements or tolerance to diseases. Our most winter hardy cultivars are still important to maintain. Timothy and cocksfoot were most stable over all locations, in contrast to perennial ryegrass, tall fescue and *festulolium*, so the necessity of regional testing of cultivars for optimal production is clear.

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Forage crop yield and nutritive value under climate change in Canada

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Abstract

Changes in forage yield and nutritive value under future climate conditions might affect the economic and environmental performance of dairy farms. Few studies, however, have simulated future forage production with a farm-scale model in areas with short growing seasons and cold winters. We assessed the potential effect of future climate change on the yield and nutritive value of a mixture of alfalfa (*Medicago sativa* L.) and timothy (*Phleum pratense* L.), pure alfalfa, and silage maize (*Zea mays* L.), with an adaptation of seeding dates, maize hybrids, and the number of cuts of perennial forage crops. Using the Integrated Farm System Model (IFSM), simulations covered three contrasting climate areas in Canada for reference (1971-2000) and future (2050-2079) periods, with three climate models and under two representative concentration pathways (RCP 4.5 and 8.5). Simulations indicated that yield of perennial forage crops would increase in all areas (+3 to +65%) with little change in forage nutritive value. Yield of silage maize would increase more in colder (+63 to +83%) than in warmer (+15%) areas, while concentrations of neutral detergent fibre (NDF) and crude protein (CP) would decrease in the colder areas only.

Keywords: forage mixture, alfalfa, timothy, maize silage, protein, fibre, IFSM

Introduction

In Canada, forage crops, fed to dairy cows mostly as silage, typically include perennial forage species like alfalfa and timothy, and annual forage species like maize. Climate change is expected to globally increase agricultural productivity in Canada (Qian *et al.*, 2016), and changes in crop nutritive value are also expected. This could affect the performance of dairy farms, but few studies have projected future forage production with a whole-farm model. The Integrated Farm System Model (IFSM) allows the simulation of forage yield and nutritive value as one of the processes of the whole-farm system, and Jégo *et al.* (2015) showed that it can be used under current climate conditions in northern regions of North America. We report in this paper the projected effect of future climate change on the yield and nutritive value of a mixture of alfalfa and timothy, pure alfalfa, and silage maize.

Materials and methods

Using IFSM (version 4.2), simulations were run on virtual farms representative of three contrasting climate areas in Canada: Central Alberta (CAB), Quebec Southwest (QSW), and Quebec East (QE). During the growing season (1 Apr. to 31 Oct.), the average temperature is currently warmer in QSW (14.0 °C) than in QE and CAB (10.6 °C), and CAB has less cumulative precipitation (398 mm) than QE (552 mm) and QSW (625 mm). Projected forage yield and nutritive value from a reference period (1971-2000) and a future period (2050-2079) were compared. Three climate models (CanESM2, CanRCM4, and HadGEM2) were used. Simulations for the future period were done under RCP 4.5 and 8.5, with respective atmospheric CO_2 concentrations of 514 and 639 μmole mol^{-1}. Long series of 300-yr synthetic weather data were generated for each climate model and future scenario (RCPs 4.5 and 8.5) using the stochastic weather generator AAFC-WG (Qian *et al.*, 2016).

Virtual farms representative of CAB, QSW, and QE grew 96, 28, and 100 ha of perennial forage crops (pure alfalfa in CAB and alfalfa-timothy mixture in QSW and QE), and 64, 36, and 25 ha of silage maize, respectively. Pure alfalfa was not fertilized. The alfalfa-timothy mixture received 100 to 120 kg of available N ha^{-1} in the form of dairy manure. Silage maize received 175, 165, and 130 kg of available N ha^{-1} in CAB, QSW, and QE, respectively, from a combination of manure and synthetic fertilizer. We considered a three-year stand life for pure alfalfa and a four-year stand life for the alfalfa-timothy mixture. Adaptation strategies for the future period were an increase in the relative maturity index of silage maize hybrids, earlier seeding dates for all crops, and more annual cuts for perennial forage crops, in accordance with the projected increase in growing degree-day (GDD) accumulation. Additional N was applied to compensate for increased crop uptake with increasing yield.

Results and discussion

Projected increases in temperature and GDD (base 5 °C) for the future period were +3.9 °C and +660 °C days under RCP4.5, and +5.2 °C and +962°C days under RCP8.5, averaged across the three virtual farms for the growing season. Cumulative precipitations increased in all virtual farms (+47 mm). However, precipitation decreased for specific months (e.g. July in CAB and Sept. in QSW and QE). Due to projected greater evapotranspiration, the precipitation deficit for the growing season increased by 57, 37, and 18 mm in CAB, QSW, and QE, respectively.

The projected climate change was favourable to crop yield in all virtual farms, but especially in the colder areas (CAB and QE; Figure 1). As expected, the yield increase was more pronounced for maize (+56%, on average) than for perennial forage crops (+27%), because maize is highly dependent on GDD accumulation. Thivierge *et al.* (2016) projected an increase in mid-summer water stress in QSW and QE for the alfalfa-timothy mixture, which would likely also occur in CAB, with less precipitation than

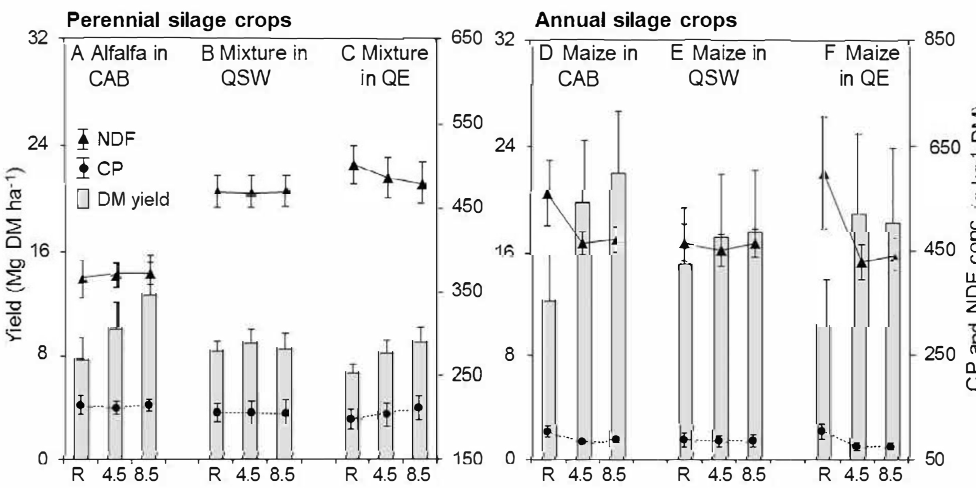

Figure 1. Yield and nutritive value (crude protein, CP; neutral detergent fibre, NDF; dry matter yield, DM) of perennial (A, B, C) and annual (D, E, F) silage crops simulated for Central Alberta (CAB), Quebec Southwest (QSW), and Quebec East (QE), for a reference period (R; 1971-2000) and a future period (2050-2079) under representative concentration pathways 4.5 and 8.5.

in the reference period in July. An increased temperature stress was also projected, especially in QSW for timothy (Thivierge *et al.*, 2016), which can explain the smaller yield increase of the alfalfa-timothy mixture in this area.

Although nutritive value was expected to decrease with projected yield increases, the CP concentration of silage maize decreased only in areas with the largest yield increase, i.e. CAB (-18%) and QE (-29%), and maize NDF concentration decreased by 16 to 27% (Figure 1D, F). For perennial forage crops, future climate did not affect nutritive value, except for a slight increase in CP concentration and decrease in NDF concentration in QE (Figure 1C). According to Thivierge *et al.* (2016), the lower nutritive value of summer cuts in future scenarios will be counterbalanced by the greater nutritive value of the first cut taken earlier and additional cuts taken at the end of the longer growing season. This is in line with the meta-analysis results from Dumont *et al.* (2015) that concluded that climate change will not affect forage nutritive value provided that farmers harvest their crops earlier under future climate conditions.

Conclusions

With adaptation of seeding dates, maize hybrids, and the number of cuts in perennial forage crops, climate change is predicted to enhance yield of perennial and annual forage crops, particularly in the colder areas. Little change in nutritive value is projected for perennial forage crops, but lower CP and NDF concentrations are projected for maize silage in colder areas. The effects of those changes in forage yield and nutritive value on the economic and environmental performance of dairy farms are currently under investigation.

Acknowledgments

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Searching for the ideal cover crop in forage maize: undersown tall fescue versus post-harvest sown Italian ryegrass

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Abstract

We compared two strategies for installing cover crops in forage maize (*Zea mays*): (1) undersown tall fescue (*Festuca arundinacea*) sown simultaneously with the maize and (2) Italian ryegrass (*Lolium multiflorum*) sown after the maize harvest. We report on data of two field trials installed in 2014 and 2015 on a sandy loam soil in Belgium. We used contrasting herbicide treatments to study weed control, the development of the undersown tall fescue and the competition with forage maize. A herbicide treatment based on tembotrione, dimethenamid-P, terbuthylazine and topramezone resulted in a good weed control and allowed an acceptable development of tall fescue without dramatic yield losses of the forage maize. With this treatment, maize yield was 92% in 2014 and 83% in 2015 compared to a reference without undersown grass. At the end of the very mild autumn in 2014 the yield of the Italian ryegrass was on the same level as that of the undersown tall fescue. On the other hand, in the cooler autumn of 2015 ryegrass yield was significantly lower than that of the undersown tall fescue. We conclude that significant potential benefits are associated to undersown tall fescue in forage maize that warrants further research into this crop combination.

Keywords: *Festuca arundinacea*, *Zea mays*, herbicides

Introduction

The 'greening' part of the 2013 reform of the EU's Common Agricultural Policy requires three actions of farmers to receive full direct payment: diversifying crops, maintaining permanent grassland and dedicating 5% of arable land to 'ecological focus areas' (European Commission, 2013). A vast majority of the farmers in Flanders use cover crops to comply with the ecological focus area. To be eligible in the perspective of ecological focus area Flemish regulation demands the cover crops to be sown before October 1st. This makes sense, as cover crops sown later than the beginning of October rarely develop well. Possible cover crops for sowing after the harvest of forage maize include Italian ryegrass (*Lolium multiflorum* L.) or winter rye (*Secale cereale* L.) (Cougnon *et al.,* 2015a). However, harvesting forage maize varieties before the end of September requires early maturing varieties, which have a lower yield potential than later maturing varieties (Swanckaert *et al.*, 2015). Undersowing grass in maize offers the opportunity to fulfill the ecological focus area without being limited by the annual deadline of October 1st. Tall fescue (*Festuca arundinacea* Schreb.) and red fescue (*Festuca rubra* L.) are species with appealing properties for undersowing in maize: due to their slow early vigour, they can be sown simultaneously with the forage maize without hampering the early development of the maize. In addition, both grass species tolerate several herbicides frequently used in maize.

The aim of this study was to compare undersowing a cover crop with sowing it after the harvest of forage maize, both in terms of maize yield as in terms of the supply of soil organic matter.

Materials and methods

We compared the yield of forage maize with and without undersown tall fescue in field trials on a sandy loam soil in Melle (Belgium), both in 2014 and 2015. The followed methodology was the same in both years and is described in detail in Cougnon *et al.* (2015b). Here we only provide a summary. At the end of April forage maize was sown at a density of 114,000 seeds ha^{-1}. With the exception of the reference plots, the maize was oversown the same day with a tall fescue mixture (Proterra Maize, Barenbrug, the Netherlands) at a density of 20 kg seeds ha^{-1} using a conventional seed drill. We applied five contrasting herbicide treatments in the 3-4 leaf stage of the maize, affecting the undersown grass differently. These were applied according to a randomized complete block design with three replicates. For the present study, we only consider the herbicide treatment based on 88g ha^{-1} tembotrione + 280 g ha^{-1} dimethenamid-P + 250 g ha^{-1} terbuthylazine + 33.6 g ha^{-1} topramezone because it allowed a good development of the undersown tall fescue and a good weed control in both years. Maize harvest took place near the end of September, at a dry matter content of around 30%. Few days after the harvest, the reference plots were rotavated and sown with Italian ryegrass at a density of 40 kg ha^{-1}. On five occasions during autumn and winter, the aboveground biomass of the undersown tall fescue and of Italian ryegrass was determined. On each plot, 1 m^2 grass was harvested manually by cutting the grass tillers just above the soil surface. The harvested biomass was rinsed with tap water to remove soil particles and was dried for 16 h at 75 °C. ANOVA compared the performance of maize and cover crops, using the *aov()* function in R.

Results and discussion

In both years, maize yield was lower with undersown grass (P=0.031 in 2014; P=0.049 in 2015) compared to a reference without undersown grass. On average the undersown grass depressed the maize yield with 14% (8.8% in 2014 and 19.3% in 2015 (Figure 1A)). The larger effect in 2015 can be explained by the better spring development of the undersown tall fescue in combination with the dry summer conditions (163.3 mm of rain in 2015 compared to average of 231 mm in June-July-August) resulting in higher levels of competition for water. Our results thus far suggest that sowing a very early maize variety, that can be harvested at maturity around mid-September and that is followed immediately by a cover crop of Italian ryegrass might be the better agronomic option. Indeed, according to the Belgian variety lists of 2014 and 2015 (Pannecoucque *et al.*, 2014) the average yield of the three earliest forage maize varieties (19,445 kg DM ha^{-1} in 2013, 19,862 kg DM ha^{-1} in 2014) was 7.1% lower compared to the three latest varieties (21,038 kg DM ha^{-1} in 2013, 21,280 kg DM ha^{-1} in 2014).

At the end of the winter of 2014-2015 (25 March) the Italian ryegrass (2,201 kg DM ha^{-1}) outyielded the undersown tall fescue (1,463 kg DM ha^{-1}) (P=0.030). In the 2015 trial on the other hand, Italian ryegrass did not overtake tall fescue. Biomass yield in mid-February 2016 was 2,631 kg DM ha^{-1} for the

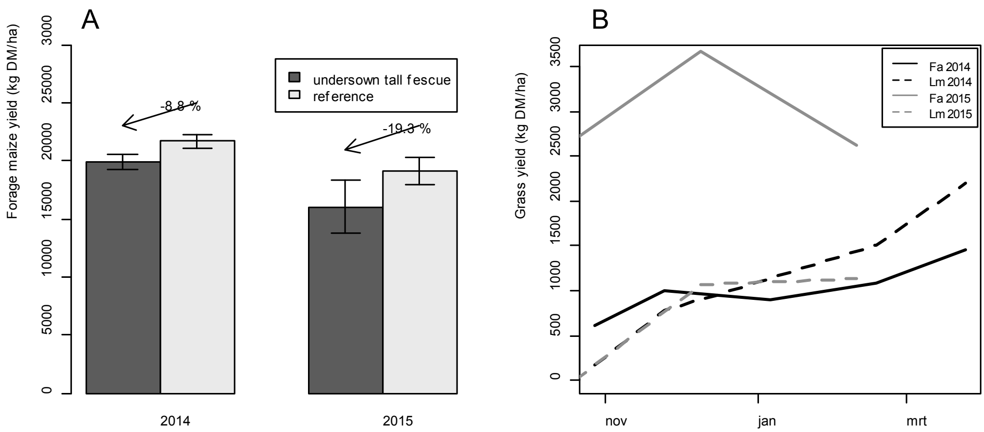

Figure 1. Field trials in 2014 and 2015 with and without undersown tall fescue. (A) Yield of forage maize; error bars denote standard errors; DM = dry matter. The arrows indicate the magnitude of the yield difference. (B) Evolution of the aboveground biomass of undersown tall fescue (Fa) or Italian ryegrass (Lm) sown after maize harvest.

undersown tall fescue versus 1,143 kg DM ha^{-1} for Italian ryegrass (P<0.001) (Figure 1B). The biomass of the undersown tall fescue was decreasing from December till February (Figure 1B). This was particularly the case in 2015 and can be explained by a growth stop of tall fescue, whereas the decay of the oldest leaves was continuing.

From the ecosystem services' point of view, undersowing tall fescue seems the better strategy. Although the standing biomass of Italian ryegass may outperform that of the undersown tall fescue at the end of the winter, we visually observed a lot more organic litter under the form of decaying leaves in the undersown tall fescue. This litter may be an important food source for the soil wood web and soil organic matter (SOM). In addition, tall fescue is known for its higher root biomass compared to ryegrass (Cougnon *et al.*, 2013). Installing the cover crop after the maize harvest further requires a soil cultivation, which stimulates SOM mineralization and increases the risk of nitrate leaching compared to the undersown tall fescue. The system of undersowing needs a further optimisation in order to restrict the competition with maize. Options are: (1) decreasing the sowing density, (2) sowing only in the maize interline zone or (3) using different species and/or genotypes.

Conclusions

Undersown tall fescue affects maize yield negatively. Based on our current data, growing an early maturing variety followed by Italian ryegrass as a cover crop provides better agronomic advantages than using a later maturing variety undersown with tall fescue. On the other hand, the ecosystem services (C sequestration) provided by the undersown tall fescue seem to outweigh the benefits associated with the Italian ryegrass sown after the maize harvest.

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Varying growth behavior of *Lolium perenne* L. clones under drought conditions and after rewatering

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Abstract

Perennial ryegrass (*Lolium perenne* L.) is one of the most important forage grass species in Europe. It is considered to be drought susceptible and will be especially affected by impacts of global climate change with expected seasonal shifts in rainfall patterns and increased frequencies of temporal drought periods. In the presented greenhouse experiment ten preselected ryegrass clones with contrasting recovery behavior after drought stress, complemented by a *Festulolium* and a meadow fescue (*Festuca pratensis* L.) clone, were investigated under optimal and limited water supply. Leaf growth rates were measured during the drought phase and after rewatering. Under increasing soil water deficit the tolerant *Lolium* clones stopped leaf growth later than the susceptible clones, but at a low growth rate each. After rewatering, most of the tolerant *Lolium* clones showed an increased growth speed within the first six days exceeding the well-watered clones and ended up with an increased leaf length after 24 days. Most susceptible *Lolium* clones showed an opposite reaction with delayed regrowth rates and decreased leaf length. The *Festulolium* and meadow fescue clone showed no enhanced growth rates and behaved more like susceptible ryegrass clones. One conclusion from the experiment was that there is only little variation for growth rate under drought stress in perennial ryegrass but recovery behaviour seems to be a promising trait for enhancing drought tolerance.

Keywords: *Lolium perenne*, drought tolerance, recovery, variation

Introduction

The effects of global climate change with increased frequencies of drought periods and shifts in rainfall patterns in Europe during growing seasons (IPCC, 2013) will have a major impact on plant production with expected yield reductions. This will also influence grassland, which can be often found on agriculturally marginal sites and especially the most important species for grassland, perennial ryegrass (*Lolium perenne* L. PR). PR is extensively used for turf and forage use due to high productivity, high cutting tolerance and high nutritional quality for livestock. On the other hand perennial ryegrass has reduced persistence under stress conditions such as drought. Thus large parts of intensively used grassland will be affected by global climate change as in most cases irrigation is not possible due to economically and technical reasons. Using natural variation in breeding new varieties with enhanced drought tolerance, which can be expected in perennial ryegrass due to a wide geographical range of origin, is one of the most promising approaches to face the upcoming problems. This paper presents the results of a perennial ryegrass greenhouse trial with intensive phenotyping during water withhold and recovery periods.

Materials and methods

Ten PR clones originating from gene bank accessions and breeding material were investigated. They were preselected for divergent drought stress response in a two-year rain-out shelter experiment in natural soil according to visual biomass scorings after drought stress (tolerant PR clones 36_21; 36_37; 81_13; 82_1; 170_30; 170_39, susceptible PR clones 108_18; 112_38; 132_4; 132_13). One representative of *Festulolium* (FEL) and meadow fescue (MF; *Festuca pratensis* L.) each, showing drought tolerance in the field, were added to the set for comparison. The material was tested for drought tolerance in a pot experiment under controlled greenhouse conditions. The experimental design was a randomized complete block with three replicates and two treatments (optimal water supply vs drought stress). Clones were planted in pots (15×15 cm) with standard potting substrate (75% black peat, 25% white peat, 9 vol. % clay granulate, 8 kg m^{-3} bentonite). At 2015-07-07 (cutting timepoint 3) all pots were filled to a maximum water content. Until 2015-08-11 (cutting timepoint 4) soil water content was reduced continuously in the drought variant. To keep the stress level comparable between all pots soil water content was measured with a TDR sensor and watering was performed to compensate for differences in evaporation. Water was withheld completely after cutting timepoint 4 (day 0) and leaf length was measured manually three times a week as the longest leaf per plant starting with day 4. Drought treatment was stopped as nearly all pots showed volumetric water content of 0% at 2015-09-08 (cutting timepoint 5; day 28). The stressed variant was rewatered by flooding the pots for 5 h. During regrowth period the water content of all pots all pots was kept at field capacity and leaf length measurements were continued until 2015-10-02 (cutting timepoint 6; day 52). The software package Plabstat version 3A (Utz, 2011) was used for ANOVA analysis, calculation of repeatabilities, adjusted means and least significant difference (LSD).

Results and discussion

In unstressed plants repeatabilities for daily growth rates ranged between 43.8 and 94.0% from day 0 to 28 (LSD5 increased from 0.89 to 11.88 cm), and between 25.0 and 80.4% from day 29 to 52 (LSD5: 1.29-11.29 cm). In the stressed variant repeatabilities for growth rates ranged between 28.2 and 61.8% (LSD5: 0.32-4.17 cm) during increasing drought stress and 0.0 to 75.0% (LSD5: 0.90-8.92 cm) during recovery period. As expected, repeatabilities were decreased in the stress variant compared with the well-watered plants, even though the trial was conducted under controlled greenhouse conditions. The genotype 132_13 was not considered for further analysis; due to poor biomass growth the stress level was different for this genotype.

Figure 1 shows the leaf growth of clones (adjusted mean of three replicates) under increasing drought stress and after recovery. With increasing soil water deficit most of the drought tolerant classified clones stopped leaf growth approx. at day 20 (36_21; 36_37; 81_13; 82_1), for most of the susceptible classified clones (108_18; 112_38; 132_4; 132_13) the growth curve flattened from day 17 on. The drought tolerant checks FEL and MF behaved like the susceptible PR clones. Total leaf length under drought stress reached between 19.7% (36_21) and 37.1% (112_38) of the well-watered control.

During the recovery period, most of the tolerant classified clones (36_21; 36_37; 81_13; 82_1; 170_39) showed an increased growth speed within the first six days compared to the well-watered control. At day 52 the well-watered clones reached leaf lengths similar to cutting timepoint 5, the stress-treated plants ended up with increased leaf lengths. This effect was already described for two other perennial ryegrass genotypes (Volaire *et al.,* 1998). Most susceptible clones (108_18; 132_4; 132_13) showed an opposite reaction with delayed regrowth rates and decreased final leaf length at day 52 compared to the control. The tolerant classified clone 170_30 behaved like the susceptible genotypes and the susceptible one 112_38 like a tolerant genotype, indicating other mechanisms underlying the drought response of these clones. FEL and MF showed a drought response like susceptible PR clones. Drought tolerance of MF and FEL is supposed to be mainly based on deep rooting (Zwicke *et al.,* 2015) in natural environments, which is disabled in a pot experiment. Regarding the biomass dry matter production during recovery phase, only the stressed variants of 36_37, 82_1, 112_38 and MF could exceed the well-watered variants (data not shown), indicating that also other yield components influence total dry matter yield during drought recovery.

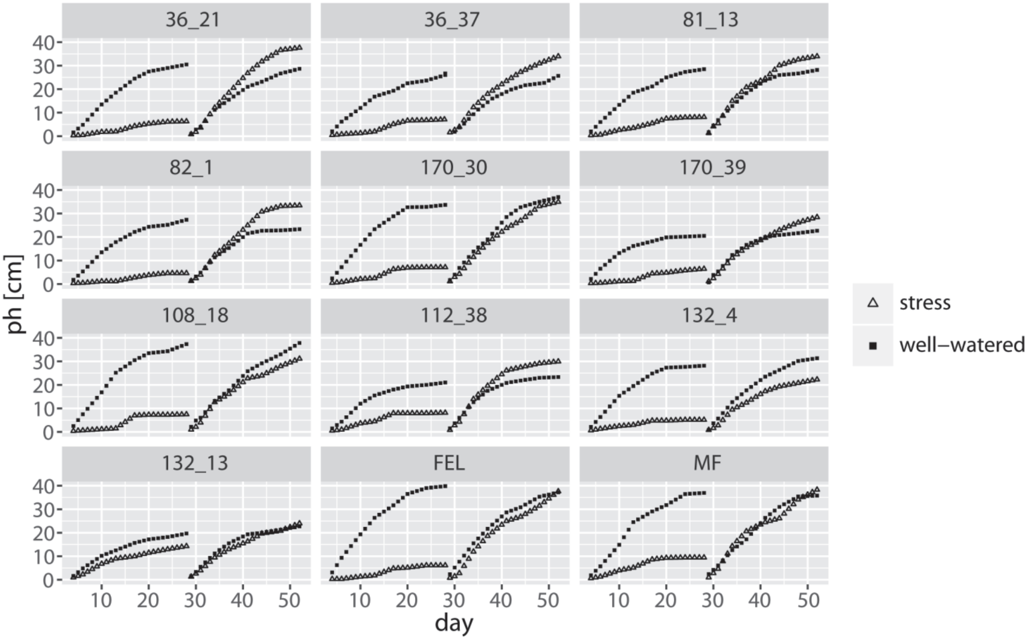

Figure 1. Adjusted means of leaf growth rates of the single perennial ryegrass genotypes under well-watered and drought stressed conditions during drought stress (day 4-28) and rewatering (day 29-52).

Conclusions

Differences in growth rates during drought stress could be detected, but all genotypes ended up with a severe reduction in biomass formation. The more promising trait for breeding drought tolerant perennial ryegrass is the selection for fast recovery after a limited time of water scarcity. This study showed that variation for recovery is present in PR. which can be used for building segregating populations to study the underlying genetic mechanisms.

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Changes in freezing tolerance and photosystem II acclimation mechanisms within a *Lolium perenne* × *Festuca pratensis* substitution series

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Abstract

Lolium perenne cultivars are widely grown for their high value forage throughout the temperate world. A limiting factor to their use is their susceptibility to winter stress. The closely related species *Festuca pratensis* is more tolerant of winter stress and can produce hybrids with *L. perenne* which combine the attributes of both parent species. In this study a seven *Festulolium* substitution lines where one of each pair of *L. perenne* chromosome was replaced with its homeologue from *F. pratensis* as well as their parental genotypes were subjected to freezing stress and their response investigated in order to locate putative genes for freezing tolerance.

Keywords: *Lolium perenne, Festuca pratensis*, cold acclimation, freezing test, chlorophyll fluorescence

Introduction

Perennial ryegrass (*Lolium perenne*) is considered an ideal grass for European agriculture however it does have constraints to its use including freezing tolerance and this can limit their productivity in areas where freezing stress is encountered (Humphreys *et al.*, 2006). Whereas *L. perenne* is susceptible to low temperature, the closely related and winter hardy species, *Festuca pratensis* (which hybridises naturally with *L. perenne),* provides significant opportunities for interspecific chromosome recombination to enhance the freezing-tolerance of *L. perenne.* Cold acclimation (CA) is a process leading to the development of freezing tolerance (FT) through a range of biological modifications including changes in the photosynthetic mechanisms. Changes in the redox state of photosystem II (PSII) caused by low temperature have been proposed as a temperature sensing mechanism for cold acclimation (Rapacz, 2004), the protection of the photosynthetic apparatus at low temperature is an important component in the increasing of FT. The aim of this study was to examine the effect of temperature on the plants FT and studying mechanisms of photosynthetic acclimation too cold by measuring the changes in PSII before and after CA. This knowledge will allow the location of putative *Festuca* delivered gene combinations to enhanced expression for freezing tolerance.

Materials and methods

The plant material used was a *L. perenne* × *F. pratensis* introgression mapping family comprising 11 plant genotypes. These included the parental genotypes and seven *L. perenne /F. pratensis* monosomic chromosome substitution lines where each of the seven chromosomes of *L. perenne* was replaced with its *F. pratensis* homeologue (Harper *et al.,* 2011). The plants were transferred into a fully controlled environmental growth room and during the first week, a temperature of +15 °C was maintained for 24 h (photoperiod 10/14 h day/night) and 300 μmol m^{-2} s^{-1} PPFD light. The plants were then cold acclimated for 4 weeks at +2/5 °C (day/night) with photoperiod and lights as above and then tested for freezing. 10 clonal tillers of each genotype for each temperature (-8, -9, -10 and -12 °C) were placed in plastic tubes and transferred to freezers where they were exposed sequentially to a rate of 1 °C h^{-1} temperature declines between +2 and -12 °C and their recovery subsequently recorded. When the decrease achieved the temperature of -8 °C a quarter of the plants were returned to the conditions of cold acclimation where they were defrosted for two hours. The temperature of the remaining plants was then successively reduced to -9, -10, and 12 °C. Defrosted plants were planted in trays of compost and transferred to a growth room (+15 °C, 10/14 h photoperiod, 300 μmol m^{-2} s^{-1} PPFD). The regrowth was estimated after three weeks using the scale where 0 = dead plant, no sign of leaf elongation; 1 = plant is surviving. The temperature having a 50% depression in regrowth score compared with control plants (LT_{50}) was estimated. Detailed studies of chlorophyll fluorescence measured with a OS-30p portable fluorometer (Opti Sciences, Hudson, USA) were used to estimate the effect of cold acclimation on the maximal quantum efficiency of photosystem II (F_v/F_m) and the changes in the ability to dissipate excess light energy non-photochemically (NPQ efficiency). Plants were dark adapted for 30 minutes prior to measurements. F_v/F_m was recorded at a saturating actinic light intensity of 3,500 μmol m^{-2} s^{-1}. All fluorescence measurements were done in 10 replications (leaves) for each plant, on the middle section of the youngest, fully expanded leaf. Measurements before CA were done at 15 °C and during the first day growing under 2 °C temperature regime. Measurements of cold acclimated plants were done under conditions of CA. The parameters calculated were: ($F_v/F_m = (F_m - F_0)/F_m$), $F_v = F_m - F_0$ and NPQ = $(F_m - F_{m'})/F_{m'}$.

Results and discussion

The *L. perenne* × *F. pratensis* interspecific hybrids show a range of freezing tolerances with chromosome 4 and chromosome 5 having the highest LT_{50} being capable of withstanding freezing temperatures in excess of -12 °C (Table 1). A similar result was shown for the *F. pratensis* parent. The studies of chlorophyll fluorescence performed prior to CA (measured at 15 and 2 °C) and after CA (at 2 °C) showed that low temperatures generally generated a decrease in F_v/F_m, but the slight changes found, were the evidence that the plants at this stage were not subject to photoinhibitory damage (Table 1). The biggest decrease in F_v/F_m was observed for chromosome 1, 2 and 3 with decreases of 8, 7 and 7% respectively. These substitution lines were characterized as cold susceptible having lower tolerance then their parental genotypes. The other cold susceptible line (chromosome 7) showed a decrease in F_v/F_m values of (6%). In frost tolerant chromosome 4 and chromosome 6 an F_v/F_m decrease after CA was 5%. Overall no significant differences in F_v/F_m values were observed between the *Lolium* and *Festuca* parents and their *Festulolium* progeny, however, post CA differences were observed in non-photochemical quenching of chlorophyll fluorescence (NPQ). Initially the reduction of temperature caused an increase in NPQ in the

Table 1. The effects of cold acclimation (CA) on LT_{50}, maximum quantum yield of PSII (F_v/F_m) and non-photochemical quenching of chlorophyll fluorescence (NPQ).[1]

Genotype	$LT_{50\ °C}$	Fv/Fm before CA	sd	Fv/Fm first hours of CA	sd	Fv/Fm after CA	sd	NPQ before CA±sd	NPQ First h of CA±sd	NPQ after CA±sd
Lp Meltra	-11.5	0.790	0.01	0.776	0.01	0.755	0.01	0.58±0.17	0.88±0.71	0.32±0.19
Fp Bf 1183	-13.6	0.789	0.01	0.767	0.01	0.753	0.01	0.70±0.13	0.45±0.19	0.66±0.31
F1	-12.7	0.790	0.01	0.785	0.02	0.754	0.01	0.89±0.22	0.66±0.31	0.57±0.34
Lp Liprior	-12.2	0.786	0.02	0.758	0.03	0.737	0.03	0.51±0.36	0.95±0.50	0.56±0.26
Chromosome 1	-9.4	0.792	0.01	0.767	0.02	0.729	0.04	0.70±0.29	0.83±0.52	0.70±0.34
Chromosome 2	-10.7	0.799	0.01	0.753	0.04	0.745	0.02	0.35±0.22	0.79±0.66	0.90±0.43
Chromosome 3	-10.4	0.791	0.01	0.769	0.01	0.732	0.03	0.85±0.45	0.39±0.23	0.52±0.36
Chromosome 4	-13.5	0.786	0.01	0.730	0.02	0.743	0.03	0.75±0.33	0.63±0.25	0.69±0.27
Chromosome 5	-13.7	0.792	0.01	0.745	0.05	0.740	0.03	0.63±0.27	0.29±0.14	0.28±0.19
Chromosome 6	-12.0	0.785	0.01	0.734	0.03	0.748	0.03	0.77±0.24	0.84±0.54	0.92±0.22
Chromosome 7	-9.0	0.778	0.01	0.736	0.03	0.730	0.03	0.92±0.26	0.71±0.53	0.82±0.26

[1] sd = standard deviation.

L. perenne parents but this later reduced during CA. In contrast, *F. pratensis* Bf 1183 had reduced NPQ during the first hours of CA but it increased after completion of acclimation. This supports previous research (Humphreys *et al*, 2006) which reported the presence of different mechanisms of energy dissipation in *Lolium multiflorum* and *F. pratensis* with the non-photochemical mechanism expressed particularly by *F. pratensis* but were not evident in *L. multiflorum*. The substitution lines showed a range of NPQ changes, in some cases with a *Festuca*-like mechanism (chromosome 3, 4 and 7) and in another the *Lolium*-like mechanism (Chromosome 1). Frost tolerance was accompanied by an increase in light energy dissipation in some progeny (chromosome 4 and 6), but was not observed in chromosome 5. The results demonstrate that the substitution of *L. perenne* chromosome 4 and chromosome 6 by its homeologue from *F. pratensis* can enhance freezing tolerance and confer improved ability to acclimate its photosynthetic apparatus to cold condition via a non-photochemical mechanism. Whilst the *Lolium* genetic background will differ for each substitution line the findings that enhanced NPQ and freezing-tolerance expression occurs when *F. pratensis* chromosome 4 is present provides consistent evidence that genes associated with enhanced freezing-tolerance are located on that chromosome (Humphreys *et al.,* 2006). During the experiment a significant (P<0.05) negative correlation (-0.61) between LT_{50} and F_v/F_m measured after CA was observed but there was no significant relationship between LT_{50} and NPQ after CA (0.388) or F_v/F_m and non-photochemical quenching after CA (-0.158).

Conclusions

The results have shown variation in frost tolerance and capability to cold-acclimate the photosystem apparatus within of *L. perenne* × *F. pratensis* substitution lines. However, the changes in Fv/Fm and NPQ do not explain the differences in frost tolerance. For chromosome 4 and 6 and increase in both parameters has been observed which suggests the presence of a non-photochemical acclimation mechanism in these lines The evidence of a role for genes found on chromosome 4 of *F. pratensis* for increasing freezing tolerance was previously reported. This paper also shows that the source for increased NPQ during CA derives from the *F. pratensis* genome and not from the *L. perenne* genome. The genes involved are likely to reside on *F. pratensis* chromosome 4. The role of genes on chromosome 6 will be the subject of further investigation.

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The North Wyke Farm Platform: the impact of ploughing, cultivations and reseeding on emissions of CO_2 and N_2O

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Abstract

The North Wyke Farm Platform was established in 2010 as a UK national capability and its remit is to research agricultural productivity and ecosystem responses to different management practices for beef and sheep production in lowland grasslands. A system based on permanent pasture was implemented on three 21 ha farmlets, each containing five catchments, in order to obtain baseline data in hydrology, nutrient cycling and productivity for 2 years from April 2011. In 2013 two catchments were reseeded with a deep rooting grass, either with or without white clover. Fluxes of CO_2 and N_2O were measured on each of the three farmlets using automatic chamber systems linked to photoacoustic analysers. Small effects of soil disturbance on emissions were measured in the three months after the intervention.

Keywords: emissions, ploughing, cultivation, reseeding, soil, disturbance

Introduction

Agricultural activities contributed to 8% of UK national greenhouse gas (GHG) emissions in 2012 (MacCarthy *et al.*, 2015). Whilst different grass cultivars are sown to improve a range of ecosystem services, disturbances to an established permanent pasture may cause greater emissions of GHG. This study investigated the short term consequences of ploughing and reseeding on emissions of CO_2 and N_2O on the North Wyke Farm Platform (NWFP).

Materials and methods

In 2013 separate automatic opaque chamber systems, each comprising twelve units, were deployed in three permanent pasture fields (Catchments 13 to 15) on the NWFP when grazed by sheep from March onwards. Continuous measurements of gross CO_2 flux were undertaken using a Licor infra-red gas analyser (LI-8100A; www.licor.com), and of N_2O using a LumaSense photoacoustic field gas monitor (INNOVA 1412i; www.lumasenseinc.com). The fields were fertilised with 40 kg N ha^{-1} on 5 March, 10 April, 7 May and 6 June applied as ammonium nitrate. Catchments 14 and 15 were sprayed with glyphosate on 27 June; ploughed on 10 July; cultivated between 18 July and 7 August when they were reseeded with a deep rooting grass *Festulolium* cv. Prior with (Catchment 14) or without (Catchment 15) white clover (*Trifolium repens* cv. AberHerald). Catchment 13 remained as a permanent pasture control and was grazed for the remainder of the grazing season.

Results and discussion

Before Catchments 14 and 15 were sprayed with glyphosate (Figure 1) the inputs of nitrogen fertiliser were evident in small 'spikes' of N_2O and CO_2 release. Reliable data was only available in Catchment 13 from 11 June, and so this has been omitted. All three catchments contained permanent pastures in this period but, whilst there are similarities in the trends in Figure 1, there are some instances when one of the systems was not working (e.g. mid May). Mean values for Catchment 14 and 15, respectively, were 111 vs 165±11.1 (kg CO_2-C ha^{-1}) and 69 vs 141±13.4 (g N_2O-N ha^{-1}).

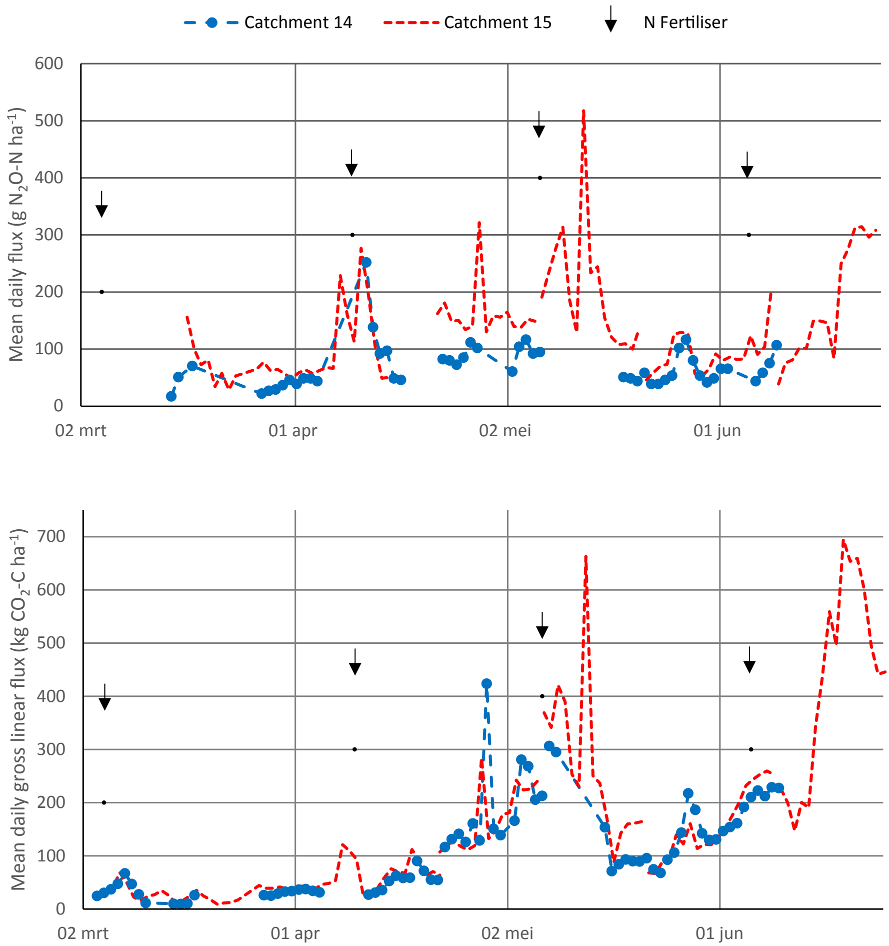

Figure 1. Mean daily flux of N_2O-N and CO_2-C on permanent pastures (4 March to 26 June).

After the spraying of glyphosate (Figure 2) the main comparisons that can be made are between the permanent pasture control (Catchment 13) and Catchment 14 because servicing requirements for the N_2O analyser in Catchment 15 meant that data was not available for some of this period. However, after spraying and during a 2-week period when the plants were dying in Catchments 14 and 15, the emissions for Catchments 13 to 15, respectively were 61, 172 and 134 (g N_2O-N ha^{-1} d^{-1}) and 228, 164 and 133 (kg CO_2-C ha^{-1} d^{-1}). Whilst the losses following ploughing and cultivation (Figure 2) appeared to be similar, the picture is somewhat complicated by loss of data at times resulting from equipment failure and resourcing issues around the requirements to move then replace the equipment before and after field operations. There is evidence that disturbance in Catchment 14 (Figure 2) resulted in greater losses of N_2O-N in September compared with the control permanent pasture (Catchment 13). Unfortunately, the high losses of CO_2-C on Catchments 14 and 15 in August coincided with a period when data was not available for Catchment 13 and so it is not possible to say that disturbance increased the CO_2-C losses as a result of the aeration and oxidation of soil organic matter.

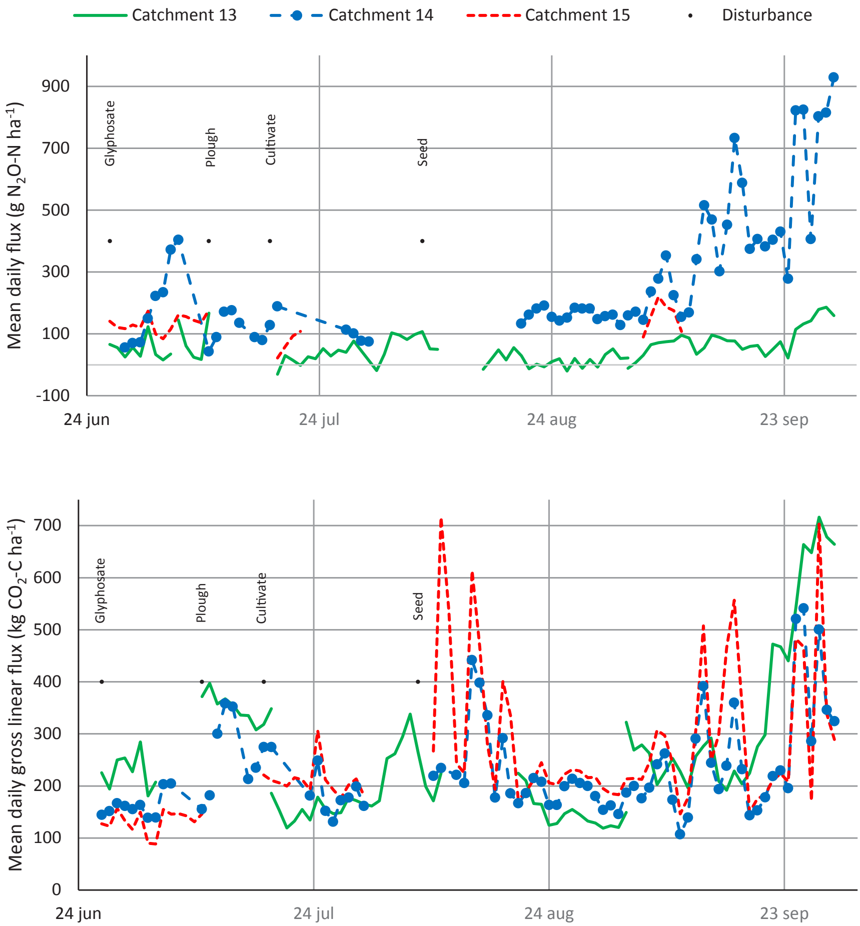

Figure 2. Mean daily flux of N_2O-N and CO_2-C on permanent pasture (Catchment 13) and reseeds (Catchments 14 and 15) from 27 June to 30 September.

Conclusions

Spraying, ploughing, cultivation and seeding were found to have small effects on the emissions of N_2O and CO_2 in a short term period. Further analysis will help to separate the effects of spatial and temporal variability, and evaluate the impact of soil heterogeneity.

Acknowledgements

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A tannin extract to mitigate methane emissions in dairy cows grazing on tropical pasture

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Abstract

Enteric methane emissions per kg of dry matter (DM) intake are usually greater in ruminants grazing on tropical grass pastures than ruminants grazing on temperate grass pastures. The aim of this work was to assess if including *Acacia mearnsii* tannin extract in the diet of dairy cows on a tropical grass-based diet could mitigate enteric methane emissions without reducing herbage intake and animal performance. The treatments were lactating dairy cows grazing on pearl millet (*Pennisetum glaucum* L.) and receiving 6 kg of concentrate feed (750 g kg^{-1} of ground corn and 250 g kg^{-1} of soybean meal) with or without 20 g kg^{-1} of *A. mearnsii* tannin extract. Twelve Holstein cows were assigned in a switch back experimental design. Each experimental period was conducted over 28 days, with a 21 days adaptation and a 7 d measurement period. Total DM intake (average 17.1 kg cow^{-1}), DM pasture intake (average 11.8 kg cow^{-1}) and milk production (average 19.2 kg cow^{-1}) were similar between treatments. Enteric methane emissions decreased by 34% (P=0.082) in cows that were supplemented with tannin extract compared to cows that were not. Dairy cows receiving *A. mearnsii* tannin extract up to 120 g^{-1} day can reduce enteric methane emissions without compromising productive performance.

Keywords: tannins, feed additives, rumen fermentation, greenhouse gas, milk production, dairy cow

Introduction

The accumulation of greenhouse gases (GHG), mainly carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O), in the atmosphere contributes to global warming. Enteric methane emissions are a major source of GHG of dairy systems. Enteric methane emissions per kg of dry matter (DM) intake in ruminants grazing on tropical grass pastures is usually greater than in ruminants grazing on temperate grass pastures, because of the quantity and profile of structural carbohydrate (Archimède *et al.*, 2011). On the other hand, relatively low levels of condensed tannins in diets (<20 g kg^{-1} DM) showed a variable effect on methane emissions by ruminants (Jayanegara *et al.*, 2012) and high levels can result in a drop in productive performance. The aim of this work was to assess if including *Acacia mearnsii* tannin extract in the diet of dairy cows on a tropical grass-based diet could mitigate enteric methane emissions without reducing herbage intake and animal performance.

Materials and methods

The work was conducted in Lages, SC, Brazil (27°47' S; 50°18' W and 920 m altitude) during autumn 2014. Twelve Holstein cows were assigned in a switch back experimental design. The treatments were lactating dairy cows grazing on pearl millet (*Pennisetum glaucum* L.) and receiving 6 kg of concentrate feed (750 g kg^{-1} of corn ground and 250 g kg^{-1} of soybean meal) with or without 20 g kg^{-1} of *A. mearnsii* tannin extract (CT; Weibull Black, Tanac S.A., Montenegro, Brazil). The grazing method was rotational grazing with a pre-grazing sward height not higher than 60 cm and a post-grazing sward height not lower than 30 cm. Experimental periods lasted for 28 days, with 21 days to adaptation and 5 days to collection period. Individual cow milk production was measured twice daily (07:00 and 16:00 h). Milk composition was determined in samples collected during milking, in the last 5 days of each period. The plasma nonesterified fatty acids (NEFA) were analysed in blood samples obtained via jugular vein in the last day of each period. Samples were stored in tubes without anticoagulant and immediately cooled down on wet ice and centrifuged, and plasma was cooled and stored at -80 °C for subsequent analysis. Concentrate intake was measured by the difference between offered and refused concentrate in the last 5 days of each period. Individual pasture intake was measured during the last 5 days by the *n*-alkane technique (Mayes *et al.*, 1986), considering the ratio of pasture C_{33} (tritriacontane) to dosed C_{32} (dotriacontane). Faecal grab samples from each cow after each milking, and pasture samples through the grazing simulation collected from each treatment were collected during the 5 days of the measurements. The faeces and pasture samples were oven-dried at 60 °C for at least 72 h and then ground through a 1-mm screen for subsequent chemical analyses. Enteric methane emissions were measured using the sulphur hexafluoride tracer technique. Data were analysed with Proc Mixed (SAS Institute 1999). Values of $P>0.10$ were considered not significant.

Results and discussion

Inclusion of *A. mearnsii* tannin extract on concentrate supplementation decreased methane enteric emissions on 34 and 30% per animal (P=0.082) and per kg of DM intake (P=0.078), respectively. Carulla *et al.* (2005), with a similar extract in sheep supplementation, also observed reductions on enteric methane emissions, attributing this effect to reductions on rumen degradation of fibre and other nutrients. Grainger *et al.* (2009) tested a similar *A. mearnsii* tannin at levels higher than those tested in this study and also observed a reduction in methane emissions, although DM intake and milk yield were negatively affected. In the present study, pasture and total DM intake were not affected by the treatments (P>0.10). The pre- and post-grazing sward heights were similar among treatments, and the pasture residual height in both treatments were greater than 60% of pre-grazing sward height, indicating a non-restrictive pasture offer (Table 1). Milk production and milk constituents were not affected by extract tannin (P>0.10), by contrast with the results obtained by Grainger *et al.* (2009). The blood NEFA content was slightly lower on the extract tannin-based treatment (P=0.012). NEFA reflects the magnitude of mobilization of fat from storage and can be used as an energetic balance indicator for dairy

Table 1. Pasture characteristics and animal performance of dairy cows grazing pearl millet (*Pennisetum glaucum* L.) and supplemented with 6 kg of concentrate feed (750 g kg^{-1} of corn ground and 250 g kg^{-1} of soybean meal) with (T) or without (WT) 20 g kg^{-1} of *Acacia mearnsii* tannin extract. [1]

	Treatments		SEM	$P > F^2$
	WT	T		
Sward height, cm				
Pre-grazing sward height	54.0	55.8	0.74	0.231
Post-grazing sward height	34.0	36.3	0.85	0.200
Animal measurements				
Pasture DM intake, kg	11.8	11.8	0.22	0.987
Concentrate DM intake, kg	5.4	5.3	0.03	0.060
Total DM intake, kg	17.2	17.1	0.23	0.852
Milk yield, kg day^{-1}	19.2	19.3	0.37	0.808
Milk fat content, g kg^{-1}	35.9	36.7	0.34	0.157
Milk protein content, g kg^{-1}	33.5	33.2	0.22	0.471
NEFA, mmol l^{-1}	0.23	0.25	0.004	0.012
CH_4 emissions				
g cow^{-1} per day	517	340	50.8	0.082
g kg^{-1} DM intake	31.4	21.9	2.67	0.078

[1] SEM = standard error of the means; DM = dry matter; NEFA = nonesterified fatty acids.

[2] Probability of a significant effect of treatments.

cows (Reist *et al.*, 2002). However, a larger trial period is required on order to determine if the difference on NEFA content found in this study could result in production or reproductive problems.

Conclusions

Dairy cows receiving *A. mearnsii* tannin extract up to 120 g day^{-1} can reduce enteric methane emissions without reductions on productive performance. Further studies with larger trial periods should be performed in order to confirm that the results obtained in this work extend throughout a lactation period.

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Effect of different renovation and weed management strategies on botanical composition and forage yield in perennial leys

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Abstract

We investigated weed management strategies in the renewal phase with the aim to reduce the need of herbicides in the subsequent years and give high forage yields in intensively harvested leys. Three field trials were established in spring 2014 located from south to north in Norway, studying the effect of glyphosate, ploughing, rotary harrowing, sowing times, cereal cover crop and selective herbicides in the new seeded ley. Botanical composition and dry matter yield were assessed in the sowing year and first ley year. Preliminary results showed that total amount of weeds and *Rumex* spp. could be reduced by cereal cover crops (not all fields) and by selective herbicides. Effect of glyphosate on *Rumex* spp. and yield varied with time of application and location. Rotary harrowing gave more weeds and reduced forage yield compared to ploughing in some fields. The forage yield was lower when sown in summer compared to spring in the northern-most field.

Keywords: grassland, renovation, weed control, *Rumex* spp.

Introduction

Undesirable plant species or weeds may reduce forage yield levels and herbage quality in leys, thus necessitating sward renewal for achieving good yields and yield quality. However, renewal results in yield loss in the seeding year, and ploughing is costly and time consuming. Extending the interval before renewal is therefore desirable, and farmers experience that weed control in the newly seeded crop leads to reduced weed infestation and less need for herbicides in the subsequent ley years in short-term leys. All farmers should practice integrated pest management (IPM) according to the Directive 2009/128/EC on sustainable use of pesticides. IPM implies use of a combination of preventive and direct measures against harmful organisms. There is, however, lack of knowledge on how to efficiently reduce weed infestation during sward renewal and optimize weed control by applying different seeding strategies. In this study, we compared different integrated weed management strategies in the renewal phase with the aim to find strategies that reduce the need of herbicides in the subsequent years and give high forage yields in intensively harvested leys. Here we present the results from the sowing year and the first harvest year.

Materials and methods

Three field trials with a split-plot design and three replicates were established in 2014 in weed infested grassland at three locations in Norway: Særheim (58 °N), Kvithamar (63 °N) and Holt (69 °N). On the main plots we compared the effect of different renewal strategies (Table 1) including fallowing (2,160 g glyphosate ha^{-1} applied on large rosettes of weeds), mouldboard ploughing (20-24 cm depth), rotary harrowing (7-10 cm depth), different sowing times and cereal cover crop (spring barley). Effect of selective herbicides in the new seeded ley (when seeded grass had 2-3 leaves and red clover (*Trifolium pratense* L.) had one coupled leaf) where studied on subplots (a=without herbicide, b=with herbicide). Herbicides were tribenuron-methyl+MCPA (3.75 g+375 g a.i. ha^{-1}) at fields seeded with grass+red clover mixture (Særheim, Kvithamar) and fluroxypyr+clopyralid+MCPA (80+40+400 g a.i. ha^{-1}) at Holt seeded with grass mixture. The percentage ground cover of weeds and crop species was assessed in spring before ploughing and in autumn after the last field operation. Botanical composition and dry matter (dried 48 h, 60 °C) yield were assessed in the sowing year and first ley year. The number of *Rumex* spp. was counted per plot at the same time. The results were analysed by variance with 'proc mixed' in SAS (SAS Institute Inc., Cary, NC, USA). Tukey-Kramer test and a significant level <0.05 were used to distinguish significant differences.

Results and discussion

Rumex longifolius DC. was present at all sites at start of the experiment. Særheim had *Rumex obtusifolius* L. in addition. Other weed species were present. Here we focus on *Rumex* spp. All renovation treatments reduced *Rumex* spp. in the first ley year compared to before start of treatments in 2014 (Table 2). After sowing, a lot of annual weeds emerged in addition to some perennial weeds at all sites (not shown). The total amount of weeds was reduced by cereal cover crops (treatment 2 vs 1) in fields at Kvithamar and Holt and by selective herbicides in all fields (not shown). The effects on *Rumex* spp. show the same tendency (Table 2). At Særheim, use of cover crop increased the number of *Rumex* spp. and total weed biomass. The forage yield in first ley year was increased by use of selective herbicide only at Særheim ($P<0.001$), while the use of cover crop did not have any effect (Figure 1).

The effect of glyphosate applied after first cut on *Rumex* spp. in the first ley year was not significant in any fields (treatment 4 vs 3, Table 2). The effect of glyphosate on total weed biomass was better (30-60% reduction in first cut of first ley year compared to mowing). The effects of glyphosate on forage yield varied with time of application and location (Figure 1). At Særheim and Holt, glyphosate in early

Table 1. Treatments on main plots in fields sown in 2014 at Særheim, Kvithamar and Holt. Treatment 6 was an extra treatment at Særheim and Holt.

Treatment no.	Time of renovation	c. 10 d before tillage	Tillage	Cereal cover crop
1	spring	no treatment	ploughing	no
2	spring	no treatment	ploughing	yes
3	summer (after 1st cut)	mowing	ploughing	no
4	summer (after 1st cut)	glyphosate	ploughing	no
5	summer (after 1st cut)	glyphosate	rotary harrow	no
6	early summer	glyphosate	ploughing	no

Table 2. Number of *Rumex* spp. (plants per 10 m^2) at start of trials in spring 2014 and at first and last (2nd or 3rd cut) cut in the first ley year in 2015 at Særheim, Kvithamar and Holt on main plots (T, cf. Table 1) and subplots (H, see text).[1]

Location	Year	Time	Treatments on main plots (T)							Herbicide (H)			T × H
			1	2	3	4	5	6	Sig.	a	b	Sig.	Sig.
Sær-heim	2014	At start	2.0	15.3	6.4	5.1	20.3	2.2	ns	9.2	7.9	ns	ns
	2015	1st cut	1.5	5.1	1.7	0.3	0.2	0.2	*	1.7	1.3	ns	ns
	2015	3rd cut	2.3	6.7	1.7	0.6	2.6	0.4	ns	2.6	2.1	ns	ns
Kvit-hamar	2014	At start	40.7	28.1	40.3	32.3	71.1	–	ns	39.2	45.8	ns	ns
	2015	1st cut	4.7	1.7	7.2	5.1	43.3	–	***	19.0	5.8	**	*
	2015	3rd cut	4.0	3.8	7.3	6.3	33.8	–	*	14.8	7.3	*	ns
Holt	2014	At start	9.2	2.5	19.2	16.7	15.0	19.2	ns	15.3	11.9	ns	ns
	2015	1st cut	6.9	2.6	1.7	1.6	7.6	3.9	ns	5.7	2.4	**	ns
	2015	2nd cut	7.4	5.1	1.7	1.2	2.7	6.5	ns	6.8	1.5	***	ns

[1] Significance levels of main effects and interaction T×H are given: ns = not significant, * = $P<0.05$, ** = $P<0.01$, *** = $P<0.001$.

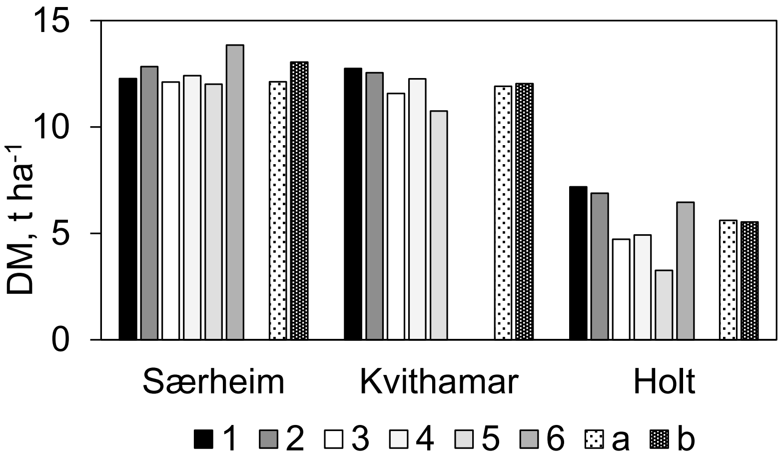

Figure 1. Forage yield in first ley year 2015 (dry matter (DM) summed over all cuts) of three field trials with various renovation methods (treatment 1-6 on main plots, see Table 1 for explanation) and without (a) or with (b) selective herbicide in new seeded ley in 2014.

summer (treatment 6) tended to give a better forage yield in first ley year than application after first cut (treatment 4). This was probably also influenced by sowing time and establishment of the new seeded crop. Rotary harrowing gave more weeds and *Rumex* spp. (Table 2), and reduced forage yield compared to ploughing at Kvithamar and Holt (not significant), but not at Særheim (Figure 1). The reason may be better performance of rotary harrowing or better effect of glyphosate before harrowing at Særheim than at the other fields. The field at Holt with shorter growing season had a low yield level, especially when seeding in summer after the first cut.

Conclusions

Ploughing and use of selective herbicides in the new seeded ley gave a good weed control. Rotary harrowing gave in two of three fields more weeds and reduced forage yield compared to ploughing. Effect of glyphosate on *Rumex* spp. and yield varied with time of application and location. The use of glyphosate in early summer gave high forage yield in first ley year. The effect of cover crop was more variable. It is important to do the operations in an optimal way taking also the weather into consideration. In areas with a short growing season this may be difficult when sowing after first cut. The long-term weed control and forage yield in the year of renovation and later years are important to take into consideration as well.

Acknowledgements

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Climate influence on an extensive managed forest steppe grassland from Romania

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Abstract

Forest steppe grassland is the most important resource of fresh fodder in the lowland area of Romania. This grassland type is used generally in an extensive manner and covers a great surface. Forest steppe grassland type has socio-economic importance for all Romanian geographical regions due to the extended surface and accessibility. In this research was considered a wide spread grassland type in Romania, respectively *Festuca valesiaca* Schleich. ex Gaudin – *Festuca rupicola* Heuff. type, *Agrostis tenuis* Sibth. subtype (after Țucra *et al.*, 1987), used by extensive grazing with sheep. The vegetation and climate data collected during six years were processed statistically having in view the correlations among vegetation and climate parameters. The obtained results show that all considered vegetation parameters were influenced statistically by at least one climatic factor considered in this research. The floristic aspects were influenced mainly by the temperature increase and the productivity by the rainfall amount fallen during the vegetation period.

Keywords: forest steppe grassland, climate, vegetation, extensive management

Introduction

The scenarios resulted from modelling in the case of steppe and forest steppe grasslands are discouraging, showing a strong decrease in the following decennia from the point of view of the aboveground biomass and the carbon:nitrogen ratio, the main factor being the dryer summers (Leary, 2008). The large scale researches regarding the evaporation during more than three decades (1951-1987), show the most significant increase of this phenomenon due to the temperature increase during the warm season in the steppe and forest steppe from southern Russia and Latvia (Golubev *et al.*, 2001). The similar trend of the evapotranspiration was noticed by Náfrádi *et al.* (2013) in the Central Europe using the paleoclimatic trends analysis. According with Mátyás (2009) in the South East Europe are extensive plain surfaces in the region placed in a broad climatic and ecological transition zone (ecotone) towards steppes and arid lands. These ecosystems are highly vulnerable to climatic changes. Other scenarios are predicting that the small areas of temperate steppe and forest steppe will be stable by remaining in the same vegetation class (Pernetta, 1995). The purpose of this paper is to summarise the six years results regarding the influence of several climatic parameters (air temperature, soil temperature, rainfall amount and air humidity) on forest steppe vegetation parameters (species richness, floristic composition, fresh fodder yield and pastoral value).

Materials and methods

The researches were carried out on forest steppe grassland, *Festuca valesiaca* Schleich. ex Gaudin – *Festuca rupicola* Heuff. type, *Agrostis tenuis* Sibth. subtype (after Țucra *et al.*, 1987) used in extensive manner by sheep grazing. The vegetation data were collected using the linear point quadrate method (Daget-Poissonet, 1971). The surveys were registered during six years (2004-2009 period), there being considered for analysis the following calculated parameters of the sward: species richness, grasses number, legumes number, other species number, grasses contribution (CS%), legumes contribution (CS%), other species contribution (CS%), fresh fodder yield (t ha^{-1}) and pastoral value (VP). The climatic factors considered were: air temperature monthly average (°C), rainfalls monthly amount (mm), soil temperature monthly average (°C) (0-20 cm depth) and air humidity monthly average (%). There were considered for analysis the average values during the months I-XII and during the vegetation period (months III-IX). The meteorological data were obtained from the Meteorological Station Oravița located at 13 km distance from the research area. The vegetation parameters and the climatic data were analysed using Bravais-Pearson correlation coefficient (r). The obtained r values were analysed using the one-tailed test ($df = n$-1, significance levels P<0.05=0.669; P<0.01=0.754; P<0.005=0.874).

Results and discussion

In Romania a great surface of land is covered with forest steppe grasslands. The climate variations certainly affect the vegetation sward, this fact being obviously noticed by the farmers. For the local economy, sustainability is important to have a predictable fodder yield and quality. The analysed correlation coefficients (r) (Table 1) for the six years of researches offered interesting results. Species richness was highly significantly correlated with the air temperature for both time intervals considered (months I-XII; r=0.92; months III-IX; r=0.77) and soil temperature during months I-XII (r=0.79). Grasses number was comprised between 3 and 6 species and it was influenced significantly by the rainfall amount fallen during the vegetation period (r=0.67). Grasses CS% ranged from 34.58 to 52.91% and was influenced by the other factors that have not influenced the grasses species number, respectively it has decreased with the increase of the air temperature (months I-XII; r=-0.68) and soil temperature (months III-IX; r=-0.68). Grasses CS% was also significantly correlated with air humidity for both time intervals considered (months I-XII; r=0.85; months III-IX; r=0.76). The number of legumes (4-11) was positively correlated with air temperature and soil temperature during the entire year (months I-XII; r=0.74; r=0.76), but their contribution (10-34.58%) was not correlated with the considered climatic data. The number of species from other botanical families ranged from 7 to 18, and their number increased highly significantly with the increase of the air temperature (months I-XII; r=0.97; months III-IX r=0.92) and significantly with the soil temperature (months I-XII; r=0.78; months III-IX; r=0.68). The legumes contribution reacted differently, respectively increases under the influence of soil temperature (months I-XII; r=0.8) and decreases with the increase of the air humidity (months I-XII; r=-0.8). The fresh fodder yield was very low and ranged from 2.5 to 7.6 t ha^{-1} and it was significantly influenced only by the rainfall amount fallen during the vegetation period (r=0.78). The pastoral value is implicitly influenced by the

Table 1. Correlation coefficients (r) among the climatic factors and vegetation parameters during 6 years.[1]

Parameters	Air temp. average (°C)		Rainfall amount sum (mm)		Soil temp. average (°C)		Air humidity average (%)	
	I-XII	III-IX	I-XII	III-IX	I-XII	III-IX	I-XII	III-IX
Species richness	0.92***	0.77**	-0.44	0.44	0.79**	0.62	-0.36	-0.45
Grasses number	0.24	-0.02	0.02	0.67*	0.14	-0.14	-0.06	0.27
Legumes number	0.74*	0.5	-0.61	0.50	0.76**	0.57	-0.62	-0.47
Other species no.	0.97***	0.92***	-0.37	0.34	0.78**	0.68*	-0.23	-0.50
Grasses CS%[2]	-0.68*	-0.55	0.52	0.27	-0.72	-0.68*	0.85**	0.76**
Legumes CS%	0.41	0.22	-0.19	0.51	0.36	0.20	-0.56	-0.29
Other species CS%	0.46	0.57	-0.56	0.13	0.60	0.81**	-0.47	-0.80**
Fresh fodder yield (t ha^{-1})	-0.17	-0.39	-0.13	0.78**	0.04	0	-0.33	0.08
Pastoral value (VP)	-0.69*	-0.54	0.42	0.10	-0.64	-0.49	0.66	0.51

[1] One-tailed test, $df = n$-1; * $P<0.05$ = 0.669; ** $P<0.01$ = 0.754; *** $P<0.005$ = 0.874.

[2] CS% = percentage contribution of species.

floristic composition. Calculated VP ranged from very low to low (VP 12.58-27.83) and it has decreased significantly with the increase of the temperature (months I-XII; r=-0.69).

The increase of the species richness under the influence of the temperature increase brings a false positive result because it is due mainly to the increase of the number of the species from other botanical families, respectively forbs and weeds. This fact is reflected mainly in the significantly decrease of the grasses contribution. This means a lower coverage of the soil during the warm season, there being created niches for different categories of opportunistic species. This evidence is also related with the decrease of the pastoral value in relationship with the increase of the air temperature.

Conclusions

The considered vegetation parameters were influenced by at least one climatic factor considered in this research. The results highlighted the great influence of the rainfall fallen during the vegetation season on the fresh fodder yield. The increase of the temperature has a greater influence on the pastoral value and floristic structure of the grassland sward, determining the decrease of the grasses contribution and a great increase of the number of the species from other botanical families. Long term researches can become able to set predictable patterns of the vegetation structure and production evolution under the influence of the climatic changes. It is difficult to assess the evolution trend of the vegetation considering the climatic pressure. Such researches must to be developed considering more many parameters of the vegetation and climate.

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Cereal cover crops in interaction with undersown red clover seed production at prolonged growing season

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Abstract

Warming climate raises a hypothesis, whether intermediate two-row barley and early spring wheat could replace the previously recommended six-row barley as cover crops during red clover seed field establishment. We sowed early red clover Varte (4n) at four rates under the cover of barley and wheat at four nitrogen treatments. The growing period of Mooni was 11 days longer, hence the seed yield of clover decreased by 20%. The respective seed yields were 388 and 316 kg ha^{-1}. The sowing rate of cover crop affected the seed yield of clover by 1.5%, nitrogen rate by 2.5%, reduction of both yielded up to 5.8% more seed. Advisable sowing rate for red clover Varte (1000 seed weight 3.15 g) under both cereals was 4 kg ha^{-1}. Neither the cover crop species and its nitrogen rate, nor the sowing rate of clover affected the 1000 seed weight and germination of clover. The monetary value of the crops was calculated. Considering the establishment cost and sale prices of cereals and clover seed, barley proved more profitable cover crop. Present-day weather allows establishment of the red clover's seed field under the cover of barley Maali and spring wheat Mooni.

Keywords: cover crop, *Trifolium pratense* L., seed yield, establishment costs, monetary value

Introduction

Red clover (*Trifolium pratense* L.) is a most important herbage legume in Estonia. An earlier recommendation was to establish its seed production fields under the early six-row barley varieties. Meanwhile the seeding and nitrogen rates of the cover crop were diminished. Recommended sowing rate for tetraploid red clover varieties is 6-12 kg ha^{-1} (Bender, 2006). Extension of the growing season caused by global warming and release of lodging-resistant spring cereal varieties raises a question about their suitability as cover crops. Neither the reductions of seeding rates of cereals nor their nitrogen fertilization are considered necessary in Norway (Aamlid and Havstad, 2011). The same authors suggest reduction of the clover's sowing rate to 2-4 kg ha^{-1}. Field trials were carried out at Estonian Crop Research Institute (ECRI) in 2013-2014, aiming to verify this advice in Estonian climatic conditions.

Materials and methods

Two field trials were established according to identical experimental design. An intermediate two-row barley variety Maali was used as a cover crop in one, an early variety of spring wheat Mooni in another. Both cereals constituted four variants: (1) seeding and nitrogen rates reduced by one-third (control); (2) seeding rates unreduced, nitrogen reduced by one-third; (3) both seeding and nitrogen rates unreduced; (4) seeding rates reduced by one-third, nitrogen unreduced. The treatments were derived from full sowing rates, viz. 500 germinable seeds m^{-2} for barley and 600 for spring wheat, 90 kg ha^{-1} of nitrogen for barley and 120 for wheat. The plots of early tetraploid red clover Varte (1000 seed weight 3.15 g) were sown across the drills of cover crops at the rates of 2, 4, 6 and 8 kg ha^{-1} of pure live seed. Randomised plots of clover were replicated four times. The trials were established to calcareous cambisol (Ko) with the following soil nutrient contents: P 179, K 162, Ca 1392 and Mg 56 mg kg^{-1}. Organic carbon content was 20 g kg^{-1}, soil pH_{KCl} 5.4. Compound fertiliser Skalsa was distributed at a rate of 400 kg ha^{-1}, i.e. P 19, K 67 kg ha^{-1} before the establishment. Nitrogen was applied as ammonium nitrate according to the experimental plan. Cover crops were sown at narrow drill space on 30 April by a seeder Fergusson, red clover on 2 May with a seeder Hege 80. The field was sprayed once against the weeds with herbicide

MCPA 750 at a rate of 1.0 l ha^{-1}. Barley ripened and was harvested on 30 July, spring wheat on 10 August. Grain yield of cereals was harvested in six replicates with a combine Hege 140. The grain quality was measured in the lab of ECRI using accepted biochemistry methods. After harvest the red clover plants could recover for 90 and 79 days, respectively till the end of growing season. In 2014, i.e. seed harvest year extreme weather prevailed at early bloom. Night frosts occurred above the stand on 24, 26, 27 and 28 June. At later stage of bloom in July the work of pollinators was favoured by warm and rather dry weather. Combine Hege 140 harvested the clover seed on 20 August. The trial situated 300 m away from 8 beehives. Statistical data analysis was performed by software AGROBASE 20^{TM}. The economic calculations were based on the input costs valid in Estonia in spring 2013, on the sale prices of grain in December 2013 depending on the quality and on red clover price in December 2014.

Results and discussion

Sowing rates of the cover crops reduced by one-third affected insignificantly the yields of barley and wheat. Diminished nitrogen fertilisation significantly reduced the grain yields and protein contents of both cereals, gluten content declined significantly in spring wheat (Table 1). As an average of the treatments red clover sown under barley yielded 388 kg ha^{-1} seed. If sown under spring wheat the yield was 316 kg ha^{-1}, i.e. 18.6% less (Table 2). Nitrogen applications to cover crops and the sowing rate of red clover had insignificant effect upon the clover's seed yield. Relative to the seed yield of clover, averaged over the four seeding rates, barley as the cover crop in nitrogen treatment no. 2 yielded by 1.5%, no. 3 by 5.8% and no. 4 by 2.5% less clover seed than did the control. The respective seed yield decrements resulting from spring wheat cultivation accounted for 7.4, 10.7 and 6.5%. Cover crop species and its nitrogen fertilisation had no effect on the germination and weight of red clover seed. Taking account of economic calculations, the seed field establishment with concurrent reduction of seeding and nitrogen rates in both cereals is reasonable (Table 3). Sowing under barley proved more profitable.

Conclusions

The trials revealed that at current duration of growing season in Estonia intermediate two-row barley varieties are preferable cover crops to red clover at seed field establishment. The seeding and nitrogen fertilisation rates of cereals should be concurrently reduced. The seed field of clover can be sown under

Table 1. Characteristics of cover crops.

Seeding rate of cereals from recommended (%)	66	100	100	66	LSD[1] 0.05
Nitrogen rate (%)	66	66	100	100	
Number of a treatment	1 (control)	2	3	4	
Barley Maali					
Plant height (cm)	72	69	75	79	2
Reproductive tillers m^{-2}	592	637	694	621	44
Yield (kg ha^{-1})	3,586	3,678	4,211	4,104	528
Protein content of grain (g kg^{-1})	90	87	97	101	4
Spring wheat Mooni					
Plant height (cm)	91	89	93	98	3
Reproductive tillers m^{-2}	430	517	654	616	38
Yield (kg ha^{-1})	3,474	3,331	3,703	3,707	211
Protein content of grain (g kg^{-1})	103	102	122	119	7
Gluten content (g kg^{-1})	218	222	279	270	21

[1] LSD = least significant difference.

Table 2. Seed yields of red clover Varte (kg ha^{-1}).

Sown, kg ha^{-1}	**Treatments (for details see Table 1)**				**LSD[1] 0.05**
Barley Maali	**1 (control)**	**2**	**3**	**4**	
2	389	373	349	387	44
4	402	397	377	390	70
6	413	411	398	392	49
8	389	388	374	383	46
Mean	398	392	375	388	
LSD 0.05	49	57	59	35	
Spring wheat Mooni					
2	316	310	290	311	56
4	325	306	312	319	50
6	340	329	319	325	82
8	366	302	283	305	73
Mean	337	312	301	315	
LSD 0.05	53	45	54	62	

[1] LSD = least significant differnce.

Table 3. Establishment costs of a seed production field, monetary value of grain and red clover seeds (€ ha^{-1}).

Sown (kg ha^{-1})	**Treatments (for details see Table 1)**							
	1 (control)		**2**		**3**		**4**	
	Cost	**Value**	**Cost**	**Value**	**Cost**	**Value**	**Cost**	**Value**
Barley Maali								
2	302	2,861	342	2,779	367	2,713	327	2,925
4	322	2,939	362	2,923	387	2,881	347	2,943
6	342	3,005	382	3,007	407	3,007	367	2,955
8	362	2,861	402	2,869	427	2,863	387	2,901
Spring wheat Mooni								
2	322	2,480	365	2,420	401	2,414	358	2,515
4	342	2,534	385	2,396	421	2,546	378	2,563
6	362	2,624	405	2,534	441	2,588	398	2,599
8	382	2,780	425	2,372	461	2,372	418	2,479

early variety of spring wheat but this will result in 20% lower clover seed yield. Advisable sowing rate for the seed production of tetraploid red clover is 4 kg pure live seed ha^{-1}.

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Semi-natural grasslands phenological evolution in western Switzerland over 21 years

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Abstract

Producing a sufficient amount of high quality fodder has become a challenge in grassland-based livestock agriculture especially in the context of climate change, due to grass growth responsiveness to climate anomalies. Phenology is therefore a useful indicator as it describes grasslands development linked to climate evolution. The aim of this study is to analyse trends in grassland phenology for different climate conditions. It is based on 21 years of grassland phenological records. Realisation dates of grass full heading stage, considered as first growth key point, have been analysed in order to evaluate inter-annual variation of spring precocity and trends in grassland earliness. Main findings are that phenological inter-annual variation has increased, revealing a higher frequency of early or late springs and that there are trends toward an earlier spring development (one to three days earlier every ten years) for temperate thermal levels. These ongoing changes underline the importance of using indicators such as phenology in grassland management.

Keywords: grassland, spring development, earliness, inter-annual variability, climate change

Introduction

Many studies (e.g. Buxton, 1996) have shown that grass nutritive value decreases during first growth with plant morphological development and ageing of tissues. Thus, phenological stage monitoring can be used as indicator of fodder nutritive value (Jeangros and Amaudruz, 2005). Since 1995, a phenological survey has been carried out in Western Switzerland. A yearly updated reference table is provided to farmers with grassland phenological stages for different thermal levels and three-day periods (Amaudruz *et al.*, 2015). A global analysis of this 21 years database has been realised in a way to assess past short-term trends of grassland and species earliness for different thermal levels in lowlands and highlands. These trends are then discussed regarding temperature evolution.

Materials and methods

From 1995 to 2015, about eighty grasslands were monitored every spring to record phenological development of ten grassland species. Plots were located in diverse climate situations ranging from 400 to 1,700 m of elevation and classified in ten thermal levels (Table 1). Firstly, the development of grasslands was analysed by considering all observed species together. For this, observed stages of the ten species were transformed into 'equivalent cocksfoot stages', namely stages that Cocksfoot (*Dactylis glomerata*) would display at the moment of observation. Standardisation was done with equations specific to each species (Jeangros and Amaudruz, 2005; Vuffray, unpublished data). The average of those standardised stages gave a global indication of grassland mean phenological stage.

According to Akaike information criterion, the best regression model was chosen between 1st, 2nd and 3rd polynomial functions to describe mean spring development of each plot (regression on mean standardised stages). To investigate phenological inter-annual variability and trends in grassland earliness, the equivalent cocksfoot stage of 'full heading' (whole spike visible on 50% of the grasses/flower visible on 10% of the forbs) was chosen, as it is the critical point at which nutritive value drastically decreases.

Equivalent cocksfoot full heading realisation dates were calculated for each plot with the regressions. Then, for each thermal level, inter-annual variation of those dates was studied globally with a Bartlett test and trends in those dates were analysed thanks to Theil-Sen trend estimator and Mann-Kendall trend test.

Secondly, to have more details, the same procedure was used to analyse individual species separately. Regression model was also chosen with Akaike information criterion (regression on observed stages, no standardisation) and full heading/flowering dates were obtained for each species of each plot. Trend analysis was carried out with Theil-Sen trend estimator.

Results and discussion

Grassland phenological development

Bartlett test indicates significant differences of variance of equivalent cocksfoot full heading dates ($P<0.05$): the variance of full heading dates during the first decade (1995-2004) is smaller than during the second decade (2005-2015) for every thermal level. As presented in Table 1, significant negative trends were found for lowland grasslands, full heading dates were two to three days earlier every ten years. Those negative trends are of the same order of magnitude than trends found in other studies (e.g. Parmesan and Yohe, 2003). On the other side, no clear trend was found for highland regions: equivalent cocksfoot full heading dates remained steady over the past 21 years.

Swiss air temperature trend (Rebetez and Reinhard, 2007) is assumed to be the main cause of grassland phenological trends observed in this study, as it is demonstrated that this parameter is a crucial explicative factor of spring earliness change (Menzel, 2003). Here, trends seemed to be influenced by altitude (non-significant in highland, negative in lowland). A study realised in the North of the Swiss Alps over 1965-2002 (Studer *et al.,* 2005) present similar negative phenological trends but they were observed at all altitude.

Species phenological development

For six out of ten observed species, phenological trends were stronger in lowland (from 'Rather hot' to 'Rather mild' levels) than in highland regions (from 'Rather Fresh' to 'Harsh' levels) and trends showed a gradual change (a gradient) of magnitude over thermal levels. Figure 1 shows this fact for three species: *Lolium perenne, Dactylis glomerata* and *Poa trivialis*. Nevertheless, for some species, gradients over thermal levels were very weak (e.g. change in trend for *P. trivialis* <0.07 day × decade^{-1} between two thermal levels). For others, trends showed a large variability around the assumed gradient (R^2<0.25), especially because of thermal levels for which trends are assessed on fewer observations (e.g. *L. perenne*

Table 1. Trends of equivalent cocksfoot full heading dates observed over 21 years (1995-2015) for different thermal levels.

Thermal levels	Altitude m.a.s.l. (indicative)	April to October mean temp. (indicative) (°C)	# of plots	Equivalent cocksfoot full heading stage mean date	Full heading date trends (day decade^{-1})	Mann-Kendall significance (*P*-value)[1]
(Rather) harsh	1000-1,250	8.0-10.0	271	5 June	steady	ns
Very fresh	900-1000	10.0-11.0	184	27 May	steady	ns
Fresh	750-900	11.0-12.0	197	19 May	-2.08	0.02
Rather fresh	650-750	12.0-13.0	210	16 May	steady	ns
Rather mild	550-650	13.0-13.5	128	14 May	-2.81	0.004
(Very) mild	<550	13.5-14.5	179	10 May	-3.20	0.0001
Rather hot / hot	<450	14.5-15.5	90	7 May	-2.04	0.11

[1] ns = not significant.

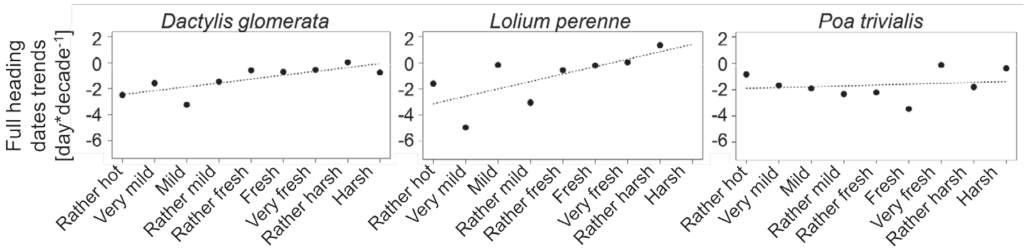

Figure 1. Trends of full heading dates for three species over 21 years (1995-2015) plotted against thermal levels. Dashed lines indicate gradual change over thermal levels.

from 'Rather hot' to 'Mild' levels*)*. It is therefore relevant to average different species by means of the 'equivalent cocksfoot stage' method, to assess more robust earliness trend.

Conclusions

According to the medium-time scale studied period (twenty-one years), changes in grassland phenological development can be outlined. Frequency of years presenting late or early spring has increased during the last decade. A trend toward earlier grassland development has been demonstrated for temperate (*i.e.* lowland) regions of Western Switzerland. Those changes highlight the importance to have indicators of grassland spring growth related to air temperature such as phenology to ensure adapted management in a context of climate change.

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Biomass yield and energy content of grass from urban roadside verges

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Abstract

Grass from urban roadside verges is a widely unused, yet potential material for energy generation systems such as (1) anaerobic digestion and (2) integrated generation of solid fuel and biogas from biomass (IFBB), which provide a press fluid for anaerobic digestion and a press cake for combustion. Urban investigation sites were established and managed in 2013 and 2014 with 4 cuts for anaerobic digestion, 2 cuts for IFBB and 8 times mulching as reference in accordance to the current management scheme. Mean annual biomass yield was 3.24, 3.33 and 5.68 t dry matter ha^{-1} for the mulching, 4-cut management and 2-cut management, respectively, with higher yield in 2014 due to favourable weather conditions. Specific methane yield of material cut on 4 dates was 232 l_N kg^{-1} volatile solids and of press fluid of material cut at 2 dates was 269 l_N kg^{-1} volatile solids. Press cake had a lower heating value of 16 MJ kg^{-1} dry matter. Gross energy output was 26.4 GJ ha^{-1} for anaerobic digestion and 84.4 GJ ha^{-1} for IFBB. To what extent fossil energy and greenhouse gas emissions might be saved by energetic utilisation of urban biomass has yet to be investigated.

Keywords: bioenergy, IFBB, city, road, green cutting

Introduction

Public authorities maintain urban roadside verges for safety and cityscape reasons. Grass is regularly cut and left in place to decay (mulching). However, authorities could use the material in energy recovery and thereby contribute to the cities' energy supply, gain revenues and support the EU waste and energy policies (European Commission, 2015). For energy recovery, two conversion technologies have been shown suitable: (1) anaerobic co-digestion (Hidaka *et al.*, 2013) and (2) integrated generation of solid fuel and biogas from biomass (IFBB; Piepenschneider *et al.*, 2015). As for anaerobic co-digestion, material of low maturity is favourable as higher degradability can be expected due to comparable low fibre concentrations. As for IFBB technique, whose main product is a combustible press cake, material of higher maturity is favourable in contrast as higher fibre concentrations regularly lead to better fuel quality. While the technical opportunities for energy recovery from material of urban roadside verges exist, it is still unclear whether the recovery is economically feasible. For economic calculation some crucial figures are missing, some of them will be given in this paper:

1. Which is the biomass yield of grass from urban roadside verges?
2. Which specific gross energy yield can be obtained by applying the two suitable conversion techniques?

Materials and methods

In the city of Kassel, which has about 200,000 inhabitants and is located in the middle of Germany, 10 investigation sites were established. These were divided in three management options: 2-cut, 4-cut and 8 times mulching as reference in accordance to the current management scheme. To measure biomass yield in 2013 and 2014, samples were taken in triplicates from a quarter of a square meter each in 5 cm height with scissors in a 12 weeks interval for the 2-cut, in a 6 weeks interval for the 4-cut and in a 3 weeks interval for the mulching management. Samples were dried at 105 °C for dry matter (DM) estimation. To detect statistically significant differences in biomass production Kruskal-Wallis test was applied. Silage was produced from the remaining biomass of the 2-cut and 4-cut managements. After six weeks ensiling minimum the material was processed with anaerobic digestion on the one hand and with IFBB technique on the other hand. For digestion tests we followed the German standard. We corrected the concentration of volatile solids applying the formula of Weißbach and Kuhla (1995). For IFBB technique we mashed the silage with warm water for 15 min before mechanical separation with a screw press. Subsequently, the C, H and N concentrations in the produced press cake were measured with an elemental analyser and the lower heating value was calculated from the higher heating value, which was generated applying the formula of Friedl *et al.* (2005), by taking the enthalpy of water vaporisation into account. Press fluid was digested in accordance to silage.

Results and discussion

In 2013, biomass yield was 4.79±3.82 t DM ha^{-1} for the 2-cut management, 2.73±1.64 t DM ha^{-1} for the 4-cut management and 2.3±1.55 t DM ha^{-1} for the mulching management. In 2014, the yield of all management options was higher (6.57±3.99, 3.94±1.86, 4.19±2.89 t DM ha^{-1} for 2-cut, 4-cut and mulching, respectively) resulting in mean annual yields of 5.68±3.90, 3.33±1.82 and 3.24±2.46 t DM ha^{-1} for 2-cut, 4-cut and mulching, respectively. The differences were not significant, neither for yield in single years nor for mean yield. Weather conditions in 2014 were favourable due to comparably high rainfall in June, July and August and warm temperatures in September and October. Although variation between sites was high as indicated by standard deviations, the mean yields of 2-cut managements are in the range of yields, which can be obtained from semi-natural grasslands (e.g. *Alopecuretum pratensis*, *Molinietum caeruleae* and *Caricetum gracilis* vegetation; Herrmann *et al.*, 2014). For weekly mowing, comparable low yields for unfertilised lawns of *Poa pratensis* (1.9 t DM ha^{-1}) and *Festuca arundinacea* (2.2 t DM ha^{-1}) have been measured (Walker *et al.*, 2007).

Specific methane yield of material cut on 4 dates was 232 l_N kg^{-1} volatile solids, of material cut on 2 dates 216 l_N kg^{-1} volatile solids and of press fluid derived from material cut on 2 dates 269 l_N kg^{-1} volatile solids. Press cake derived from 2-cut material had a lower heating value of 16 MJ kg^{-1} dry matter. Comparing a 4-cut management with subsequent anaerobic digestion on the on hand and a 2-cut management with subsequent IFBB procedure on the other hand, the measured figures indicate a gross energy output of 26.4 GJ ha^{-1} for anaerobic digestion and 84.4 GJ ha^{-1} for IFBB (Figure 1).

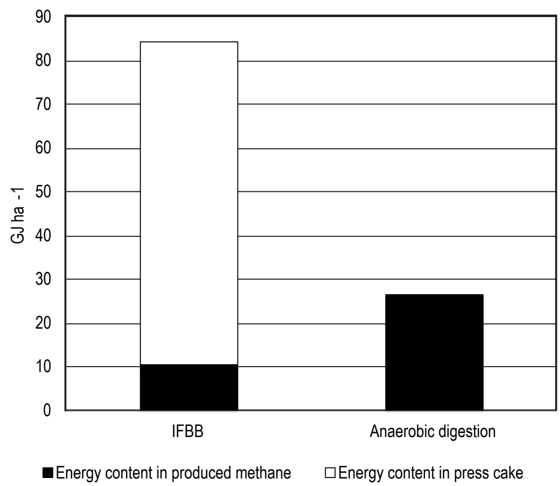

Figure 1. Energy recovery per hectare: integrated generation of solid fuel and biogas from biomass (IFBB) and common anaerobic digestion in comparison. Calculations for recovery with IFBB are based on data of the 2-cut management and for recovery by anaerobic digestion on data of the 4-cut management.

If this difference holds true when taking the energy effort along the value chain into account has to remain open for urban environments. To clarify the life cycle, special emphasis has to be placed on modelling harvesting strategies as well as mass flows within this special ecosystem.

Conclusions

The biomass yield of grass from urban roadside verges is within the range of semi-natural grassland. Energetic conversion with IFBB technique results in a gross energy output of 84.4 GJ ha^{-1}, which is about three times as high as gross energy output applying anaerobic digestion. Life cycle analysis adapted to urban environments should be followed up.

Acknowledgements

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Spatial and temporal synchronicity in Norwegian lamb weaning weights

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Abstract

We investigated correlations between time series per county for mean lamb autumn weaning weights 1992-2012. Information on 2.2 million lambs from 11 counties was used; means per county and year, corrected using a general linear fixed model, and pairwise correlations were estimated between the counties. Clearly, there are similarities in the between-year patterns between counties, especially neighbouring areas but also on a wider scale: we found high correlations between time-series. Mechanisms are not clear: summer weather factors such as temperature and precipitation are likely involved, but winter conditions may also contribute.

Keywords: sheep, weaning weight, weather conditions, time-series

Background

Norwegian sheep husbandry generally includes ≈90 days of free-range grazing, mostly on mountain pastures; most of the lamb body growth occurs in this period. Anecdotal evidence suggest that lambs will be of high weights in the same years on a regional scale; e.g. abattoirs will often report 'good' or 'bad' years for lamb weights for areas as large as all of south-eastern or northern Norway.

Materials and methods

We used data from the Norwegian Sheep Recording Scheme, choosing lambs born in spring that weighed 15-75 kg at weaning (age 95-180 days), from flocks present each year 1992-2012 with ≥40 lambs each year. There was sufficient data to analyse 11 of the 19 Norwegian counties (Figure 1). After editing, 700 flocks, with 2,233,881 lambs remained. We used a general linear fixed model to estimate corrected (lsmeans) means for weaning weight for county×year. Means were corrected for lamb age (covariate), and class variables sex (male, female), age of dam (1-8 years), litter size (1, 2, 3, ≥4), breed (short-tailed Spæl, or crossbred Norwegian White), flock, and year (21 years). We calculated Pearson correlations between county time-series of corrected lamb weights.

Results

Mean weaning weight was 43.7 kg at mean age 139 days, whilst mean dam age was 3.1 years and mean litter size 2.3 lambs. Corrected means per county and year are shown in Figure 2, pairwise correlations between the county time series in Table 1. In total, 18 of the 55 correlations were ≥0.7; only one was <0 (between counties 11-Rogaland and 19-Troms). There are in general high correlations between counties, with the exception of the northernmost, 19-Troms, which seems to vary independently of the others. The correlation analysis indicates two clusters of neighbouring counties: one in the south: 4-Hedmark, 5-Oppland and 6-Buskerud; the other with 12-Hordaland and, going north, 14-Sogn and Fjordane, 15-Møre and Romsdal and 16-Sør-Trøndelag. Breaking the pattern of highest correlations between close counties, 5-Oppland is quite well correlated (>0.62) to all other counties but Troms.

Discussion

Why the synchronicity? We expect Moran effects (Moran, 1953), with the correlated weights being caused by common environmental variables. Weather conditions are likely important, mainly acting indirectly through quantity and quality of the rangeland pasture vegetation, perhaps also through direct

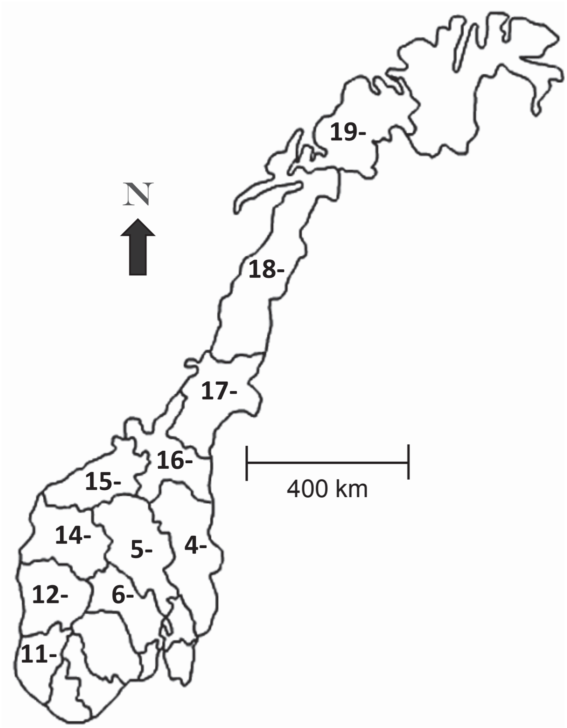

Figure 1. Codes for the Norwegian counties in the study. Names of the counties, see Table 1.

Table 1. Correlation matrix, pairwise between lamb weaning weight time series (1992-2012) for the 11 counties included in the analysis.[1]

	5-Oppland	6-Buskerud	11-Rogaland	12-Hordaland	14-Sogn and Fjordane	15-Møre and Romsdal	16-Sør-Tøndelag	17-Nord-Trøndelag	18-Nordland	19-Troms
4-Hedmark	**0.78**	**0.75**	0.51	0.46	0.53	0.56	0.63	**0.78**	0.59	0.24
5-Oppland		**0.87**	**0.70**	**0.71**	0.66	**0.77**	**0.82**	**0.72**	0.62	0.22
6-Buskerud			0.58	0.61	0.64	0.63	0.63	0.69	0.56	0.16
11-Rogaland				0.68	0.59	**0.72**	0.65	0.48	0.44	-0.03
12-Hordaland					**0.93**	**0.88**	**0.76**	0.54	0.64	0.18
14-Sogn and Fjordane						**0.89**	**0.74**	0.58	0.61	0.09
15-Møre and Romsdal							**0.85**	0.68	**0.71**	0.22
16-Sør-Trøndelag								0.67	0.68	0.23
17-Nord-Trøndelag									**0.72**	0.40
18-Nordland										0.68

[1] Correlation coefficient in bold if ≥0.7 and underlined if P>0.01 (H_0).

effects on thermoregulation and behaviour, but conditions during summer seem to have quite area-specific (local) effects (Steinheim *et al.*, 2004), making the high large-scale synchronicities surprising. Large-scale climatic effects (measured as the NAO Index) in winter have been shown to affect domestic sheep in the same way as red deer in western Norway (Mysterud *et al.*, 2001), likely through effects on the wintering conditions for the pastures both on-farm and on the rangelands. Growth conditions for forage plants the year before lambs are born could also be important, through quality and quantity of winter

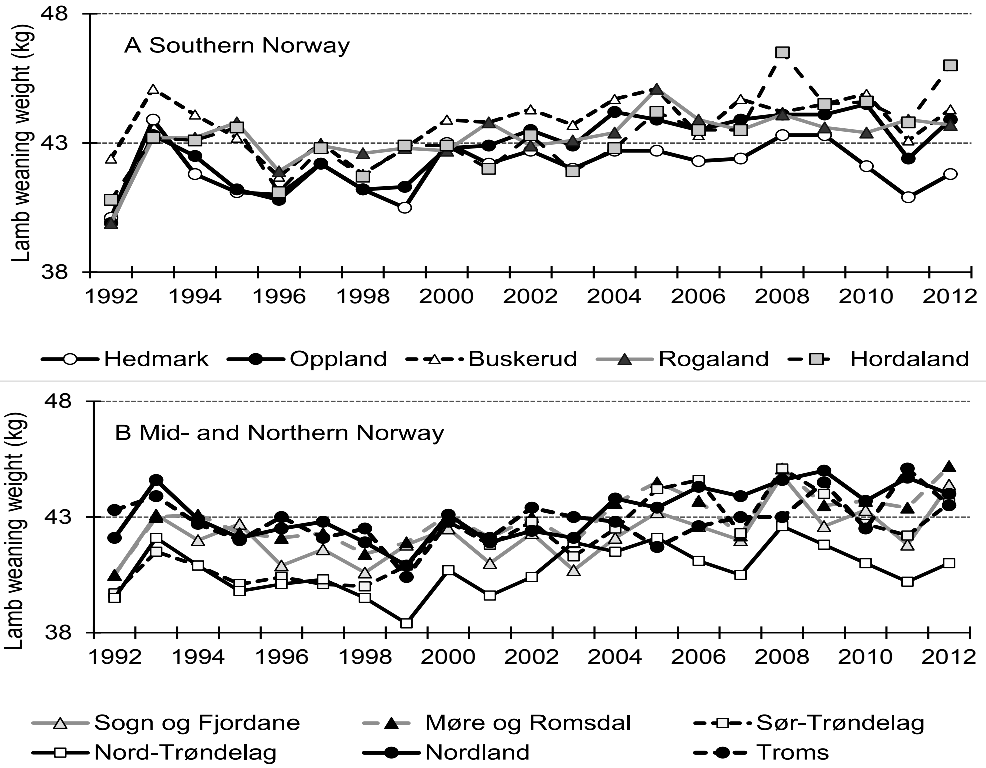

Figure 2. Corrected (least square) means for lamb weaning weight in autumn, for county by year. Corrected for lamb age, sex, litter size, breed and dam age.

forage. Finally, environmental effects may cross generations: the ewe's environment during previous years may later affect her reproductive performance and thus lambs (Steinheim *et al.*, 2002).

Conclusions

Understanding patterns of covariation is challenging; this preliminary study strongly indicates that among-years variation is not at all independent between counties. Further analyses should focus on obtaining relevant information on environmental factors, and then employ methods such as factor- and principal component analyses to obtain and test specific hypotheses.

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Farming and research – working together to develop grassland varieties resilient to water stress to mitigate and adapt to climate change

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Abstract

Extreme weather events like flooding and drought are becoming more prevalent as the effects of climate change begin to impact. To increase resilience to these, grass and clover breeders at IBERS are working to develop plants with stronger, deeper root systems and improved root-soil interactions leading to greater water-use-efficiency and improved soil hydrology. Using participatory research and working with industry partners, eight commercial development farms were selected to cover a range of soil types, geographical areas and livestock sectors (ruminant and mono-gastric) across the UK. In Autumn 2014 and Spring 2015: two 1 ha areas of single variety grass leys: *Festulolium* (cv AberNiche) and hybrid ryegrass (cv AberEve) were established to accord with each farm crop rotation and managed using their standard farm practice. The grasses were selected to form the baseline for equivalent evaluations of novel grass and clovers selected as fit-for-purpose for inclusion later in the project and for developing strategies to deliver best practice. Using a multi-disciplined approach the project SUREROOT employs two BBSRC National Capability Facilities, and the eight Commercial development farms to assess the performance of novel grass and clover populations either independently or grown as mixtures to see whether changes in individual plant design, when reproduced at the field scale, can provide a significant environmental service. This paper focusses on the role of the Commercial Development Farmers in the assessment of the field performance of novel grasses and clovers designed to withstand extreme weather events and to mitigate their worst scenarios under diverse UK livestock management systems.

Keywords: water stress, new grass cultivars, field performance, Commercial Development Farmers

Introduction

In the wet winter of 2013-2014, 50,000 hectares of the UK were flooded, resulting in an estimated £25 million of crop losses. *Festulolium* (ryegrass/fescue species) hybrids, with large deep root systems have been developed as a potential new tool for landowners to mitigate flooding. In pilot studies, *Festulolium* root-soil interactions were claimed to have instigated a change in soil structure leading to a 51% reduction in surface runoff compared to a perennial ryegrass variety (MacLeod *et al.*, 2013). Building on this work, the 'SUREROOT' project is a joint BBSRC and industry funded project aimed at developing improved rooting systems in grasses and clovers for sustainable livestock systems and for ecosystem service. The project combines researchers from two BBSRC Institutes and the farming industry through a retail supply chain network. Together, the aim is to increase resilience to extreme weather events by breeding grasses and clovers with better rooting structures, to improve water-use-efficiency, and to change soil hydrology. The protocols and research model were developed to evaluate the field performance of novel grasses and clovers and to optimise the benefits of the early involvement of the UK commercial livestock sector through participatory research (McCalman *et al.*, 2013).

Materials and methods

IBERS and the processors in a retail supply chain developed a detailed selection matrix to encompass all relevant attributes necessary for the selection of the eight Commercial development farms (CDFs)

These included monogastric and ruminant livestock farms with a diverse range of farming systems and crop rotations. The farmers were identified to cover a range of soil types, geographical regions and farm types including free range turkeys, chickens and pigs and beef, sheep and dairy enterprises. In addition to the physical characteristics of the divergent farm systems, farmers needed to be 'electronically literate', undertake farm level monitoring, be keen input their ideas to the practical application of the project outcomes and be willing to share their experiences with others.

In Autumn 2014 and Spring 2015: two paired equally-sized areas of single variety grass leys: *Festulolium* (cv. AberNiche) and hybrid ryegrass (cv. AberEve) were established to fit with each farm rotation and managed, using their standard farm practice. They form the baseline within a pilot phase to facilitate the methodology and further evaluations to be undertaken by the farmers later in the project and for developing strategies to deliver best practice. Key to the study is the participation of the farmers who record basic information (rainfall, soil temperature and any field operations) on a monthly basis and record more detailed observations on a quarterly basis, such as visual assessment of soil structure, rooting depth and earthworm numbers.

Results

To date, there has been a high level of active engagement by the farmers through the use of a participatory approach which connects them directly with the data being generated for the project. Farmers were very pro-active in the selection of field sites, using their long-term farm knowledge and expertise combined with an understanding of the needs of the research project. The participatory CDF farmers are working together with industry sector partners, bringing their sector perspective to link across all the livestock supply chains. Farmers have been taking interim measurements to give baseline information but, most importantly at this stage of the project, are learning by doing through kinaesthetic learning. The roles of the grasses in the differing farm systems range from high quality silage for the dairy farmer to resilience to turkey grazing for the poultry farms. Using information gathered to date, pilot individual case studies have been circulated to the industry supply chain members to create awareness and cascade out the potential of these novel grasses in real and diverse commercial farming situations.

Discussion

Key to the study is the active participation of the farmers involved. Although the grass variety evaluation on the commercial farms is, at this stage, not yet 'best practice' it engages farmers with the new variety development and engenders a feeling of ownership with an opportunity for robust feedback to researchers. The range of farm systems and soil types does not offer replicated scientific output but allows the opportunity to assess the suitability at a practical level of the varieties in a range of contexts. This approach has been achieved during the pilot phase to actively involve the farmers in the project whilst developing their skills to understand the processes involved in participatory research projects. There is an acknowledgement there are a wide range of mechanisms to enable research outputs to be part of shaping of innovative agriculture systems at the farm level (Gal *et al.*, 2010). The SUREROOT project seeks to encompass both reductionist research and farmer participation at an early stage in the research process. This will help facilitate the rate of adoption across the farming community and to ensure that the research developments are fit for purpose.

Conclusions

Establishing participatory farmer activity, by ensuring farmers are actively-engaged in developing the methodology used on their own farms, was found to facilitate optimal integration with the science whilst ensuring farmers feel part of the project. It ensures that the research is relevant to them as end-users of the scientific innovation. The aim of early inclusion of the commercial sector representing all areas of UK livestock agriculture was to enable a faster route to impact by providing a direct link between research and commercial practice. During the time-course of the project, outcomes from precision plant breeding technologies at IBERS and from field-based studies should provide UK agriculture with new options to better combat the climatic extremes that are likely to be encountered.

Acknowledgements

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Ash and mineral content of grasses for combustion

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Abstract

Plant biomass will be an alternative to fossil fuels in the future. However, biomass production from grasses faces challenges related to the cultivation of grass with a particular chemical content. Ash content is an important indicator of the quality of fuels, because higher ash content causes problems to the automation of the combustion process. The productivity and chemical content of grass are largely influenced by fertilizers, their types, and doses. In this research, compared to wood, ash content in grass biomass was high (4.99-7.02%), and slightly higher in festulolium (up to 7.78%). This suggests that it would be useful to produce pellets from grass biomass mixing it with wood, since this would reduce ash content. Moreover, it was found that increased potassium and phosphorus contents decreased the ash melting temperature.

Keywords: grass, fertilizers, ash, lignin, phosphorus, potassium

Introduction

Energy crops are alternative heat sources for biofuel production in many countries (Adamovics *et al.*, 2009; Bridgeman *et al.*, 2008). There are several parameters that affect the quality of solid biofuels, such as water content, lignin, cellulose, ash. Lignin and cellulose have a high heating value because of the high content of carbon in lignin (about 64%) (Heitner *et al.*, 2010; Kaltschmitt *et al.*, 2010). Compactness of the biofuel depends on the lignin content in energy crops (Alaru *et al.*, 2009). The enrichment of potassium, sulphur and chlorine in ash is very harmful as it causes fouling, slagging, and high temperature corrosion in the furnace; in addition, if aerosols are not collected in the ash separator, they may cause air pollution (Platace and Adamovics, 2012). Potassium is the dominant source of alkaline metals in most biomass fuels (Wei *et al.*, 2005). Fuel-bound nitrogen is responsible for most nitrogen oxide (NOx) emissions produced from biomass combustion. A lower nitrogen content in the fuel should lead to lower NOx emissions.

The aim of this research was to evaluate the ash, lignin, potassium, phosphorus and nitrogen content in grass depending on the type and dose of fertilizer to identify the grass biomass suitabililty for biofuel production.

Materials and methods

Field trials (2013-2014) were conducted on sod-calcareous soil (*Hypocutani-Hypocalcic Luvisol*) (pH_{KCl} 6.7; available P – 52 mg kg^{-1}, K – 128 mg kg^{-1}; organic matter – 24-28 g kg^{-1} of soil) in Latvia. The following energy crops were studied for their fuel (pellet) production: festulolium (×*Festulolium pabulare*), meadow fescue (*Festuca pratensis* Huds.), and timothy (*Phleum pratense* L.). Doses of fertilizers were: N0P0K0 (0 kg N ha^{-1}, 0 kg P_2O_5 ha^{-1}, 0 kg K_20 ha^{-1}), F – background N0P80K120 (0 kg N ha^{-1}, 80 kg P_2O_5 ha^{-1}, 120 kg K_20 ha^{-1}), F+N30, F+N60, F+N90, F+$N120_{(60+60)}$, F+$N150_{(75+75)}$, F+$N180_{(90+90)}$ kg ha^{-1}, vermicompost – 10 t ha^{-1}. Swards were cut once per vegetation period in September. Harvested biomass was weighed and analysed for dry matter content and for chemical composition using the ISO standard methods. The experimental data were statistically analysed using two-way analysis of variance with 'grass species' and 'fertiliser' as factors; the difference among means was detected by LSD at the 0.05 probability level (Excel for Windows 2003).

Results and discussion

The results showed that biomass of festulolium, timothy and meadow fescue had high ash content compared to other fuels like wood or pellets. In timothy biomass, it was slightly lower: 4.99-7.02%; in meadow fescue: 6.05-6.88%; in festulolium, it was higher: 6.57-7.78%. The study indicated the highest ash content in festulolium biomass without fertilizer N0P0K0 (control): 7.78±0.06%; in timothy biomass – 7.02±0.11%; and in meadow fescue fertilised with F+N180 $_{(90+90)}$: 6.88±0.10%. The lowest ash content in festulolium biomass was found in crops treated with F+N180 $_{(90+90)}$: 6.57±0.11%; in timothy applied with F+N150 $_{(75+75)}$: 4.99±0.10%; and in meadow fescue treated with fertilizer F+N30: 6.05±0.08%. The lowest ash content was found in fertilized grass (6.33%), followed by samples of non-fertilized grass (6.93%), and plants treated with vermicompost (6.50%). This suggests that fertilizers do not have an effect on ash content in grass (Figure 1).

Lignin content makes 3.96-4.54% in festulolium, 8.11-8.31% in timothy, and 3.53-4.07% in meadow fescue. Among all grass plants, the lowest lignin content was found in the samples of non-fertilised plants (5.38%), followed by fertilised plants (5.48%), and plants treated with vermicompost (5.50%). It can be concluded that the fertilizer did not influence the lignin content in plants. The results showed that lignin content in the timothy, meadow fescue and festulolium biomass is lower than in other energy crops. Lignin content in crop residues and perennial grasses typically ranges between 12 and 14%. Our results showed that compared to woody biomass, a lower lignin content can require more energy to pelletize grass biomass.

Phosphorus content in timothy, festulolium and meadow fescue did not differ notably and fluctuated between 0.20 and 0.26%. The highest phosphorus content was found using fertilizer P_2O_5 – 80, K_2O – 120 (kg ha^{-1}) (background) – 0.23-0.25%; the lowest phosphorus content was found using fertilizer F+N30 – up to 0.20%. The average phosphorus content in the festulolium, timothy and meadow fescue biomass depending on fertilizer type and amount varied within 0.20-0.25%. The lowest average indicators were recorded for timothy with vermicompost (0.20%) and for meadow fescue and festulolium without fertilizer (0.23%).

Nitrogen content in timothy, festulolium and meadow fescue depending on fertilizer type and amount varied from 0.69 to 1.04%. The lowest indicators were found for festulolium without fertilizer (0.69%) and for timothy and meadow fescue with P_2O_5 – 80, K_2O – 120 (main fertilizer) – 0.93 and 0.69%, respectively. This suggests that additional nitrogen fertilizer increases the nitrogen (N) content in biomass.

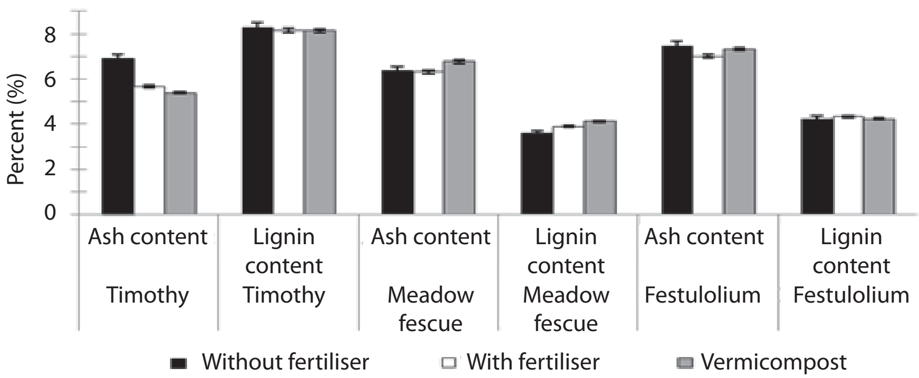

Figure 1. Average ash and lignin content in grass depending on fertilizer.

Potassium content in timothy, festulolium and meadow fescue was the lowest for timothy supplied with F+N180$_{(90+90)}$ (1.75%), for festulolium supplied with F+N90 (1.83%), and for meadow fescue supplied with F+N30 (1.37%). An average potassium content in festulolium without fertilizer accounted for 1.89%, with fertilizer 2.02%, and with vermicompost 1.84%; in timothy: 0.25, 0.83, and 0.78%; in meadow fescue: 1.53, 1.61, and 1.62%, respectively. An overly high content of K and P (which reduces ash melting temperature) causes corrosion of boiler elements. According to Aho and Ferrer (2011), such problems can be avoided if biomass is burned together with sawdust and woodchips.

Conclusions

Use of fertilizers did not influence the ash content in the grasses. Ash content in grass biomass was high, whereas lignin content was low, which suggests that it would be useful to make pellets from grass biomass mixed with wood for heat production.

The lowest ash content was found for grass biomass treated with vermicompost. The lowest ash content was recorded for the samples of festulolium and meadow fescue treated with N fertilizer, whereas the lowest ash content was recorded for timothy treated with vermicompost. The lowest ash content among the grasses was in timothy (5.91%), festulolium (7.15%), and meadow fescue (6.38%).

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New cultivars needed to ensure survival of perennial ryegrass across the northern region

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Abstract

Perennial ryegrass (*Lolium perenne* L.) is currently grown in the maritime regions of the Nordic countries but it will become a promising option further north with prolonged growing seasons and milder winters. The restricted current genetic diversity found in the region may though impede future cultivar development. As part of a Nordic Public Private Partnership on pre-breeding of perennial ryegrass, variety trials were established in Estonia, Finland, Iceland, Norway and Sweden encompassing 22 cultivars of diverse origin. The primary aim was to observe winter survival and biomass production potential over three harvest years. Significant differences in total yield over the three-year period were observed between varieties but strong genotype × environment interactions were found emphasising the importance of developing cultivars with specific adaptation, particularly to the more marginal regions.

Keywords: *Lolium perenne*, genetic resources, yield, winter survival

Introduction

Perennial forage crops at northern latitudes are limited by a short and moderately cool growing season. Projected climate change will result in a warmer and longer growing season, because of earlier spring and later autumn, thus giving increased biomass production potential. The potential gain could though be offset by factors affecting overwintering of the species grown in the region. Perennial ryegrass (*Lolium perenne* L.) is currently grown in the maritime regions of the Nordic countries but cannot be reliably grown in inland parts and north of 60°N. With its good regrowth capacity and superior feed quality, it will become a promising option further north in a warmer climate. It has been postulated that perennial ryegrass has inadequate growth cessation in autumn to build up sufficient winter hardiness (Østrem *et al.*, 2014) but recent cultivar trials have demonstrated substantial variation in environmental conditions in the Nordic region manifested by significant G×E interactions for biomass production, regrowth capacity in autumn and winter survival (Østrem *et al.*, 2015). Efficient ways must be found to incorporate new genetic variation to enhance future cultivar development. The aim of the current study was to analyse the pattern of adaptation for perennial ryegrass cultivars of diverse origin grown at contrasting sites across the Nordic and Baltic region for use in further cultivar development and here we focus on dry matter yields.

Materials and methods

Field trials with 22 cultivars of perennial ryegrass were established in spring and summer 2012 at Korpa, Iceland (62°09'N, 21°45'W), Bjørke, Norway (60°45'N, 11°12'E), Lännäs, Sweden (63°08'N, 17°43'E), Jokioinen, Finland (60°80'N, 23°47'E) and Jõgeva, Estonia (58°45'N, 26°24'E). The cultivars originated from Norway: 1. 'Einar' (4×), 2. Fagerlin (2×). 3. 'Falk' (4×), 4. Fjaler (4×), 5. Trygve (4×); Sweden: 6. SW Birger (4×), 7. SW Gunne (2×), 8. SW Irene (2×), 9. SW Leia (4×), 10. SW Svea (2×); Finland: 11. Riikka (2×); Estonia: 12. Raidi (2×), 13. Raite (4×); Latvia: 14. Spidola (4×); Netherlands: 15. Premium (2×); Germany: 16. Ivana (2×), 17. Pionero (4×); Switzerland: 18. Arvella (2×), 19. Arvicola (4×), 20. Cavia (2×), 21. Salamandra (4×); Canada: 22. Norlea (2×). Plot size ranged between 7 and 12 m^2 and the seed rate was 30 kg ha^{-1}. Plots were harvested two to four times depending on site in each of the following three harvest years at a cutting height of 5 cm and dry matter yields were measured in a standard manner. The experiments were set up in an alpha design with three replicates and five blocks within each replicate. ANOVA of total dry matter yield over harvest years, separating linear and quadratic effects, was first carried out for each site followed by AMMI analysis on predicted values derived from REML analysis in order to evaluate the adaptability and stability of the cultivars in the given environments. All analyses were carried out using GenStat® Release 18.1 (VSN International Ltd, 2015).

Results and discussion

Highest overall yields across years were obtained in Sweden, followed by Finland, Norway, Estonia and Iceland but the yield varied significantly between years at all locations (Table 1 and 2). Cultivar yield varied significantly at all locations and significant interactions between cultivars and years were observed at all locations except Norway. The residual variation was substantially higher in Finland compared to the other locations, making the results from that location somewhat dubious. In the AMMI analysis each location×year was treated as a separate environment since the pattern of C×Y varied across locations (Table 2) and the two first principal components captured around 71% of the total G×E (only PC1 presented here). The analysis shows that cultivars from Switzerland (no. 18-21) and Netherlands and Germany (no. 15-16) were less productive in the test environments compared to cultivars originating from the Nordic/Baltic region (Figure 1). They were also less stable than the other group, particularly the Swiss cultivars, which all are classified as early maturing. PC2 further separated 18 and 20 from 19 and 21 (results not shown). This group of cultivars showed specific adaptability to the more favourable environments. The Nordic/Baltic group was generally more stable, of which SW Birger (6) was highest yielding overall, but there was also considerable variation among them indicating that there is room for

Table 1. Mean total dry matter yield (t ha^{-1}) of 22 cultivars grown in the Nordic/Baltic region.

Year	Estonia	Finland	Iceland	Norway	Sweden
2013	6.65	8.10	4.79	7.89	11.74
2014	5.41	10.38	3.99	5.40	13.52
2015	5.73	10.86	5.16	6.73	7.83
mean	5.93	9.78	4.65	6.67	11.03
s.e.d.	0.158	0.471	0.212	0.159	0.193

Table 2. ANOVA for total dry matter (MS×10^{3}) across years for 22 cultivars (C) grown at five different locations in the Nordic/Baltic region.[1]

Source	d.f.	Estonia		Finland		Iceland		Norway		Sweden	
Rep	2	0.32		29.33		1.04		2.34		2.30	
Y1	1	2.81	**	251.65	**	4.38		44.27	**	506.00	***
Y2	1	2.66	**	3.59		42.61	**	160.82	***	612.00	***
Error a	4	0.82		7.32		1.49		1.23		0.83	
Cultivar	21	1.12	***	11.48	***	1.16	***	2.03	***	3.19	***
C×Y1	21	0.50	*	4.09	**	0.96	***	0.21		0.53	
C×Y2	21	2.19	***	5.89	***	0.34	***	0.20		1.54	***
Error b	126	0.26		1.70		0.13		0.14		0.41	

[1] Y1 = linear contrast over years (Yr1-Yr3); Y2 = quadratic contrast (Yr1-2*(Yr2)+Yr3).

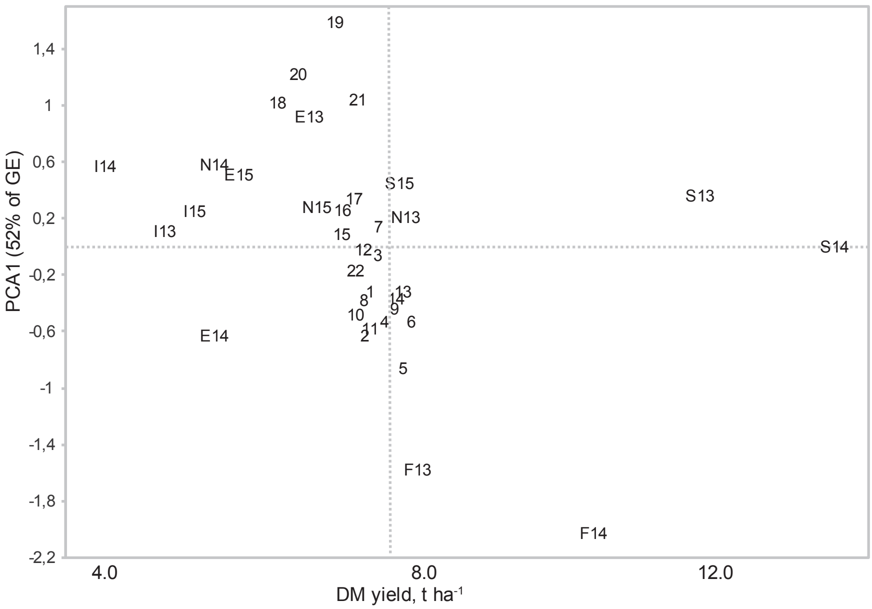

Figure 1. Biplot of 22 cultivars (numbers in italics, see designation given in text) and 15 environments (Location (E=Estonia; F=Finland; I=Iceland; N=Norway; S=Sweden) × Year (13=2013; 14=2014; 15=2015)) for mean dry matter yield (t ha^{-1}) and AMMI PC1 score.

further selection for specific adaptability, especially to the poorer environments. These results are in line with those obtained in previous studies (e.g. Østrem *et al.*, 2015), where significant G×E interactions were observed, and the next step is to elucidate whether the observed variation can be explained by particular environmental factors.

Conclusions

The study revealed highly significant G×E interactions and emphasises the importance of developing cultivars of specific adaptability, particularly to the marginal regions.

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Integrated effects of higher temperatures, acid substrate and heavy metals on red clover

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Abstract

The ability of plants to adjust to the changing climate and environmental conditions is currently becoming one of the most relevant scientific issues and has an undoubted practical significance. The aim of the present work was to study the integrated effects of substratum acidity and heavy metals (copper and cadmium) on the adaptation of red clover *(Trifolium pratense* L.) cultivar 'Liepsna' at different ambient temperatures in the phytotrone complex. Changes in plant height and shoot biomass were indicators of plant adaptation to unfavourable environmental factors. We identified the number of clover inflorescences, concentration of nitrogen and sulphur in dry matter (DM) of clover. At temperatures above (27-20 °C) clover produced a lower shoot biomass and was more sensitive to the effect of pollutants and adapted less to this effect than clover growing at optimal temperatures (21-17 °C). Higher temperature enhanced sulphur accumulation in clover biomass. Copper increased shoot biomass of clover, acted as a fertilizer at optimal temperature. Copper decreased clover DM at higher temperature.

Keywords: *Trifolium pratense*, heavy metals, substrate acidity, higher temperature

Introduction

Environmental pollution with heavy metals or acid rain has become an important factor that determines plant growth, development and productivity (Hoffmann and Persons, 1997). Such stress causes changes in physiological processes, and the consequences of the effect are determined by plant species, the outcome of the exposition is determined by plant species, variety, exposition time or strength (Alexieva *et al.,* 2003). Most heavy metals are necessary for plants, however, their excess inhibits growth and development, suppresses photosynthesis, synthesis of photosynthetic pigments, metabolism and other processes. Acidification of field and forest soils is an undesirable phenomenon, since agricultural crop yields decline in acid soils (Tong GuanHe, 2005). The objective of the present study was to investigate physiological response of red clover to the exposure to differentiated and complex acid substrate and cadmium and copper ions and to explore plant adaptive capacity to adjust to these stressors at optimal and higher tempertures.

Materials and methods

Experiments were done at the *LCARF* Institute phytotrone complex where we investigated the effects of substrate acidity and heavy metals on red clover (*Trifolium pratense* L.) cv. 'Liepsna' at different ambient temperatures. For this experiment we chose one of the most toxic heavy metals cadmium and copper, which is attributed to trace elements. The plants were grown in peat substrate in 5 l pots, in three replications. Each pot was sown with 30 seeds of red clover. The emerged seedlings were singled leaving 10 per pot. Clover was grown in a greenhouse for 45 days at 20 °C. At 4-5 leaf stage the plants were transferred to phytochambers to induce flowering and were kept for 35 days at 4 °C at 8 h photoperiod. After flowering induction the plants were transferred for 40 days to phytotron chambers (in one chamber the temperature during the daytime was 21 °C, at night 17 °C (optimal temperature), in the other chamber 27 ºC during the daytime and 20 ºC at night (high temperature), photoperiod 16 h, source of light SON-T Agro (PHILIPS) lamps. In period16-40 days the experiment with heavy metals and acidity substrate was done in two stages – the adaptation and the main at optimal and at high trmperatures (Table 1). For adaptation tests acid concentration was 6 ml H_2SO_4 l^{-1} water, the concentration of cadmium ions was

Table 1. Biomass, content of nitrogen, sulphur in dry matter (DM) and number of inflorescences of clover as affected by exposure to contaminants and temperature.

Treat-ment[1,2]	Optimal temperature (21-17 °C)					High temperature (27-20 °C)				
	Nitrogen (N) %, in DM	Sulphur (S) %, in DM	Increase in height in final test (cm)	DM biomass g pot^{-1}	Inflorescences pot^{-1}	Nitro-gen (N) %, in DM	Sulphur (S) %, in DM	Increase in height in final test (cm)	DM biomass g pot^{-1}	Inflorescences pot^{-1}
W+W	2.56	0.28	7.1	13.2	19	2.43	0.47	2.3	8.4	17
W+A	2.59	0.69	4.3	12.7	21	2.34	0.75	3.1	9.5	15
W+Cu	2.41	0.31	9.1	14.6	22	2.80	0.54	1.2	8.2	14
W+Cd	2.83	0.39	6.0	12.1	19	2.60	0.47	1.3	8.7	21
A+W	2.54	0.61	7.0	11.7	16	2.72	0.81	2.1	6.5	11
A+A	3.00	0.63	5.1	10.8	15	2.13	0.95	1.3	7.4	14
A+Cu	2.36	1.00	9.0	10.8	8	2.86	1.02	0.8	6.5	7
A+Cd	2.69	0.82	6.0	9.9	10	2.57	0.93	2.1	7.5	12
Cu+W	2.79	0.35	6.2	12.9	17	2.67	0.47	2.8	9.7	15
Cu+A	2.38	0.79	7.5	14.4	23	2.73	0.62	2.3	7.0	7
Cu+Cu	2.44	0.36	12.5	15.2	23	2.61	0.54	2.0	6.0	12
Cu+Cd	2.71	0.43	10.2	12.5	17	2.66	0.42	3.1	9.4	16
Cd+W	2.48	0.34	7.0	13.6	17	2.55	0.54	0.4	5.6	8
Cd+A	2.66	0.56	11.2	16.0	22	2.47	0.64	5.3	7.8	7
Cd+Cu	2.54	0.36	11.1	15.3	26	2.91	0.64	1.4	7.3	9
Cd+Cd	2.70	0.44	6.7	12.8	18	2.74	0.50	0.5	7.0	12
$LSD_{0.05}$	0.24	0.05	3.8	1.6	5.1	0.22	0.04	2.1	1.2	4.8

[1] The first letter: mode of exposure during the adaptation stage. The second letter: during the main stage. W: plants irrigated by water. A: exposed to 6 ml H_2SO_4 up to one litre diluted water solution.

[2] Cd: exposed to 0.16 mM $CdSO_4{\cdot}8\ H_2O$ solution; Cu: during adaptation stage irrigated with 2 mM $CuSO_4{\cdot}5\ H_2O$ solution. During the main stage – irrigated with 4 mM $CuSO_4{\cdot}5\ H_2O$ solution.

0.16 mM $3CdSO_4{\cdot}8H_2O$ (0,123g l^{-1}), and the concentration of copper ions was 2 mM $CuSO_4{\cdot}5H_2O$ (0,499g l^{-1}). Before sowing, peat substrate of slightly acid reaction (pH 6-6.5) was irrigated with these solutions. Each pot received 0.5 l solution. The pH level of the acid-irrigated substrate was 4.0. Adaptation stage lasted for 10 days. The acid concentration for the main exposure was 6 ml H_2SO_4 l^{-1} water, the concentration of cadmium was 0.16 mM $CdSO_4{\cdot}8H_2O$ (0.123g l^{-1}), the concentration of copper was 4 mM $CuSO_4{\cdot}5H_2O$ (0.998g l^{-1}). Each pot received 0.5 l of the solution. The ions of cadmium and copper did not change substrate acidity, whereas the pH of the substrate irrigated with acid solution decreased (pH 3.3-3.8). The control plants were irrigated with tap water. After the main exposure which lasted for 14 days, we measured plant height and overground biomass and carried out analysis. The number of flowers per pot was identified, as well as nitrogen and sulphur concentration in dry matter of clover biomass. The data were processed using ANOVA.

Results and discussion

Before the first (adaptation) stage clover was 33-35 cm in height, at the beginning of bud formation growth stage. Having exposed to contaminants, after adaptation stage, which lasted for 10 days, clover height was uneven. Only water-irrigated clover at optimal temperature grew up to 39-41 cm, and the clover plants exposed to 2 mM copper sulphate solution were 44-46 cm in height. The height of plants exposed to cadmium ions or acid at optimal or higher temperatures changed inappreciably during the adaptation period. After the main stage clover flowered abundantly at optimal temperature, the plants were taller than those that grew at higher temperature and were at the end of flowering stage. Red clover like other plants and living organisms, when exposed to a complex of environmental factors, respond respectively and adjust. Optimal temperature for red clover to grow is 20-23 °C. Temperatures above optimal and those above 30 °C had a greater negative effect on root growth rather than on stem growth (Frame *et al.,* 1998). In our experiments higher temperatures exerted a negative impact on clover growth and adaptation. At optimal temperature the highest increase in plant height was obtained when copper sulphate had been used during the adaptation and the main stage or during the first stage the plants had been exposed to cadmium ions and during the second stage to acid or copper ions. This is also confirmed by the dry matter data of clover overground biomass (Table 1). In these treatments biomass was significantly higher than in the check treatment where clover was irrigated by water. At higher temperature, significantly more biomass was accumulated only in one case when in adaptation stage the plants were exposed to copper ions and in the main stage the plants were irrigated by water. Biomass significantly declined, unlike at lower temperature, when in adaptation and main stages clover was exposed to copper ions. Our findings suggest that clover that grew at higher temperature produced less overground biomass than that grown at optimal temperature, and contaminants reduced overground biomass of clover. The clover that grew at optimal temperature formed more inflorescences than the clover that grew at higher temperature. Heavy metal salts with acid (A+Cu, Cu+A, Cd+A) had the greatest inhibiting effect on inflorescence formation.

Higher contents of sulphur accumulated in the stems and leaves of clover plants that grew at higher temperature, whereas nitrogen contents were similar to those identified in the plants grown at optimal temperature. The ratio of nitrogen to sulphur in the biomass was higher when clover grew at optimal temperature.

Conclusions

Red clover plants that grow at higher temperature (27-20 °C) produce less overground biomass, are more sensitive to exposure to contaminants and adapt less easily to such effects than do plants that grow at optimal temperature (21-17 °C). Higher temperature promotes sulphur accumulation in clover.

Copper stimulates accumulation of clover overground biomass and at optimal temperature acts as a fertilizer. At higher temperature copper reduces biomass.

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Contribution to C-sequestration by leys in arable rotation during a 60 year long-term trial in southeast Norway

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Abstract

It is well recognized that leys maintain higher soil organic carbon (SOC) than rotations with only annual crops; yet more precise quantification is needed. Archived soil samples taken in spring 1953 from the long-term rotational trial at Ås, and samples taken in autumn 2012, all at 0-20 cm depth, were analysed for total SOC content. During the 60 year interval, plots were cultivated in rotation with either spring cereals only, or with 2 or 4 years ley out of 6 years. There was a large variation between plots in the original carbon content. The total C loss was proportional to the initial SOC content (about 0.32 of initial SOC). In addition, cereals accelerated SOC loss. Compared to cereals only, the 2 year and 4 year ley rotations abated SOC loss with about 1.0 to 1.3 (SE ~ 0.13) ton CO_2 (ha and year with ley)$^{-1}$ in the top 20 cm of the soil profile. There was a substantial cereal yield increase correlated to SOC content, at least partly related to a better soil structure, in the post-effect year.

Keywords: soil organic carbon, crop rotation, cereal, ley, long-term trial

Introduction

The ability of leys to sequester carbon and maintain a high soil organic carbon (SOC) content could be an important contribution to mitigating the radiation forcing emission from agriculture. When leys are in rotation with annual crops, the increased SOC content can provide several other services such as, for example, reduction of some soil-borne pathogens and pests, and a better soil structure and stability, thus maintaining or improving soil fertility while reducing soil erosion and related problems, as e.g. eutrophication of fresh water by phosphorus. We used a 60 year old rotational trial at the Norwegian University of Life Sciences to study the effect of leys versus continuous cereals on the SOC and on the yield of a spring cereal crop. We expected leys to increase SOC, but this work was aimed at further quantification. Since the field lies on a relatively flat and superficially homogenous area, we expected differences prior to the start of the experiment to be negligible, compared to those imposed by the rotational treatments. We also hypothesized that there would be a diminishing benefit of increasing the number of years with ley from 2 to 4 out of a 6-course rotation with spring cereals.

Materials and methods

A long-term trial, with four 6-course rotations, was established on a silty clay loam of marine origin (20-25% clay) in spring 1953, in combination with four fertilizer management (low, moderate, moderate with some animal manure, the last treatment varied between rotations). The trial was located at Ås (59.40°N, 10.47°E), in a moderately humid and relatively cold environments (normal 1961-1990 precipitation 785 mm and average annual temperature 5.3 °C). All courses were present each year and replicated twice, making altogether 4×4×6×2 = 192 plots in a randomized split-plot design (Uhlen *et al.*, 1994).

Archived soil samples taken plot wise in the plough layer (0-20 cm) in spring 1953 and soil samples taken in autumn 2012 were analysed for total C and N content. Selected plots were also sampled for water retention curves, water infiltration, aggregate distribution and aggregate stability to rain. Post effects on the yield of a spring oats crop were measured in 2014 on all plots.

Here we present some preliminary results with focus on SOC and three rotations: (C6) only cereals; (C4) 4 y cereals and 2 y leys; (C2) 2 y cereals and 4 y leys. All cereals were spring cultivars, and leys were a mixture of timothy (*Phleum pratensis* L.) and red clover (*Trifolium pratense* L.) undersown in the spring wheat preceding the first year ley. In the regression analysis, available data from all treatments were used, unless otherwise specified. We found only 168 samples from 1953 in the soil archive. Missing samples were almost orthogonally distributed between treatments.

Results and discussion

Contrary to our expectation, there was a large variation in the SOC content at the start of the long-term experiment. The SOC distribution was relatively random, yet on average plots of treatment C2 had higher SOC content than those of C4 and C6 (average 3.9, 3.69 and 3.72 g C (100 g soil)$^{-1}$ respectively, P=0.10). Half of the variation in 2012 was still explained by the initial differences 60 years earlier (Figure 1A).

During the 60 years of the incubation trial there was a loss of SOC proportional to the initial amount (lost SOC = initial SOC × 0.32 (SE 0.05). The effect of the rotation could be accounted by an additive model: compared to the C2 rotation, the SOC loss on C6 and C4 was additionally 0.51g and 0.31 g C (100 g soil)$^{-1}$ (SE 0.035), respectively (Figure 1B). This is in agreement with Riley and Bakkegard (2007) who observed a similar loss of SOC in SE Norway during the last decades.

There was also a statistically significant effect, but moderate in size, of moderate/normal chemical N application on SOC (+0.07 g (100 g soil)$^{-1}$, SE 0.04) compared to very low N application, and an effect of 10 ton animal manure (ha y)$^{-1}$ in combination with chemical fertilization (0.09 g C (100 g soil)$^{-1}$, SE 0.04)) over the 60 year period. The effect of fertilization was additive.

C4 and C2, leys were discontinued during a whole rotational period during the 1970s. Taking this into consideration, leys were present in respectively 18 and 36 years out of 60. The ploughing depth was kept at 20 cm during the whole period, and the average bulk density measured in 2014 was about 1.3 g cm^{-3}. Based on these data, the average effect of one year of ley on the SOC content, compared to spring cereals, was 1.0 and 1.3 (SE ~ 0.13) ton CO_2 ha^{-1} for the C4 and C2 rotation respectively. At this stage we are still investigating whether the data really support the hypothesis that leys lasting for a longer period (4 years) are more effective than short (2 years) leys in preserving SOC. A confounding factor could be soil texture, which varied greatly between plots.

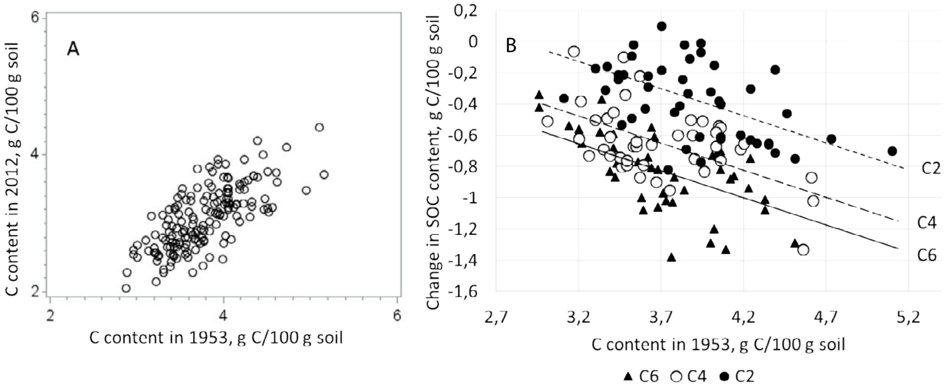

Figure 1. (A) Soil organic carbon (SOC) content in 2012 plotted against the SOC content in 1953; (B) Effect of Initial SOC content and crop rotation on the amount of SOC lost during 60 years. Lines show the results of the statistical model. Treatments are explained in the text.

The cultivated cereal area in Norway is about 300 thousand ha. Most of the cereal yield is used for feeding animals, and a large share of it is fed to cattle. At present, the share of concentrate used in cattle feeding is high, and there is room for reducing it. If each year a third of the present cereal area was cultivated with leys, and the CO_2 sequestration was as effective as in this clay loam, the SOC conservation effect would correspond to about 100 thousand tons CO_2, which is almost a fifth of the energy-related CO_2 emission from the whole agricultural sector (550 thousand tons per year), but only a minor amount compared to the 2.6 and 1.7 million tons of CO_2 equivalent emitted as CH_4 and N_2O from Norwegian agriculture (https://www.ssb.no/statistikkbanken). This study gives also an indication of the potential SOC loss if a cultivated meadow is converted to cereal. It should be noticed that the soil texture is also an important factor controlling SOC decay rate (e.g. Frøseth and Bleken 2015), and the decline is faster on coarser soil.

In addition to the positive effect of C sequestration was the enhancement of the oats yield in the post-effect year, which in C2 was 32% greater than in the rotation with cereals only, C6. The grain yield was positively correlated to SOC and there was an additional positive effect of rotation (C2 > C4 > C6 after accounting for SOC content), but not of animal manure (data not shown). A better soil structure could contribute to explain the positive after-effect of leys on yield (data not shown).

Conclusions

Omission of leys from cereal rotations in clay loamy soils in SE Norway reduces the amount of C sequestered in the soil. This amount is sizeable compared to CO_2 emissions from the use of energy by the agricultural sector, but small compared to the global warming effect of other GHG emissions from agriculture. However, there is also a considerable loss in grain yield per acreage related to the loss in SOC and rotational effects when leys are omitted from cereal rotations.

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Annual yields of intensively managed grassland mixtures only slightly affected by experimental drought events

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Abstract

We evaluated the effects of experimentally imposed drought on intensively managed grassland communities comprising four species (*Lolium perenne* L., *Cichorium intybus* L., *Trifolium repens* L., *Trifolium pratense* L.). A summer drought period of nine weeks with complete exclusion of precipitation was simulated in a common field experiment at two sites, Reckenholz (Zurich, Switzerland), and Wexford (Ireland). The drought induced severe stress at Reckenholz and extreme stress at Wexford. The short-term effects of drought reduced an individual harvest by 48% at Reckenholz, and 85% at Wexford. The yield advantage from mixing species could compensate for this short-term drought impairment under severe drought at Reckenholz. Aggregated across harvests, the annual yield of mixtures under drought exceeded the average annual yield of the rainfed monocultures. For annual yield, the benefit due to mixtures (33% increase) was substantially greater in magnitude than the impact of drought (5% reduction) in our experiment. These results illustrate the high potential for multi-species mixtures to compensate for drought-induced yield losses.

Keywords: drought, grassland yield, grass mixture

Introduction

Climate change is predicted to result in increased climate variability. The combined effects of increased variability in precipitation and the amount of precipitation per event, e.g. prolonged periods of drought or waterlogging can result in reduced yields in grassland systems (Swemmer *et al.*, 2007). In experiments with four-species mixtures in intensively managed conditions, grass-legume combinations consistently yielded more than predicted from monoculture yields (Finn *et al.,* 2013); however, there have been relatively few tests of whether such an advantage of multi-species mixtures remains evident under environmental stress, e.g. drought conditions. Although we can expect a short-term effect of drought on yields, key knowledge gaps also remain about the extent to which annual yields of intensively managed temperate grasslands are affected, and we investigate these here.

Materials and methods

A common field experiment to manipulate precipitation was established at two sites. Sites were in Switzerland at Reckenholz (near Zurich), and in Ireland at Wexford. We selected the following four forage species based on the factorial combination of their specific functional traits related to rooting depth and manner of nitrogen (N) acquisition: a shallow-rooted non-legume (*Lolium perenne* L.), a deep-rooted non-legume (*Cichorium intybus* L.), a shallow-rooted legume (*Trifolium repens* L.), and a deep-rooted legume (*Trifolium pratense* L.). Using these four species, plots (5×3 m) were established in monocultures and mixtures that were sown with equal proportions of the four species. Monocultures and mixtures were established as control treatment under ambient rainfed conditions and as drought treatment in which a summer drought event of nine weeks was simulated at both sites using rainout shelters. The annual yields comprised six harvests at Reckenholz and five at Wexford; there were two regrowth periods during the drought treatment, and one post-drought harvest. All plots of a site received the same amount of mineral N fertiliser: 200 kg N ha^{-1} $year^{-1}$ at Reckenholz and 130 kg N ha^{-1} $year^{-1}$ at

Wexford (see Hofer *et al.*, in press for details). Changes in aboveground biomass per species between the rainfed control and drought treatment were analysed based on the natural logarithm of a response ratio.

Results and discussion

Based on measurements of the soil water status, the induced drought stress was considered to be severe at Reckenholz and extreme at Wexford. Annual yield of the species in monoculture, aggregated across harvests before, during and after the drought, was on average only decreased by 5% under drought as compared to rainfed conditions (Table 1). Growing all four species in mixture resulted in substantial overyielding in annual yield at both sites and in both the control and drought treatments (Table 1). Averaged across the drought and control treatments at both sites, the mixture benefit expressed as a proportion of the average monoculture yield was 33%.

We previously reported that aboveground biomass yield of the monocultures was significantly impaired in the second harvest that occurred during the drought treatment at these sites (Hofer *et al.,* 2014), and when the drought stress was strongest. In the second harvest period during the simulated drought, the mean reduction (compared to a control) was 48% at Reckenholz, and 85% at Wexford, and the different species varied in their drought resistance. Remarkably, overyielding of the severely drought-stressed mixtures at Reckenholz was so strong that mixtures close in composition to the equi-proportional mixture reached the same yield as the average of monocultures under rainfed control conditions. Under extreme drought at Wexford, growth almost ceased in monocultures and mixtures at this time (Hofer *et al.*, 2014).

Grasslands of low to medium management intensities have been shown to be relatively resilient after drought, within the same growing season (e.g. Gilgen and Buchmann, 2009). However, less is known about the effects of drought on more intensive systems, despite their practical and economic improtance. For example, drought events impose greater economic losses on intensively managed compared to less intensively or extensively managed grassland (Finger *et al.*, 2013). Our results illustrate the nature and extent of drought in intensively managed grassland systems. At the scale of individual harvests, there can be severe effects of drought on yield, although these are probably mediated by the pre-drought levels of soil moisture, as well as the moisture retention properties of the soils once drought is underway. At the scale of total annual yield, however, the yield benefit due to mixtures (33% increase in annual yield) was substantially greater than the effect of drought (5% reduction in annual yield) in our experiment.

Table 1. Annual aboveground biomass yield (kg DM ha^{-1} year^{-1}, mean ±1 SE, n=3) of the four species in monoculture, the average of these four species, the equi-proportional mixture of the four species and the mixture benefit under rainfed control and drought conditions at Reckenholz and Wexford. Also shown is the percentage change of annual yield (% CAB) due to drought. Statistical inference is based on the natural log transformed data.

	Reckenholz				**Wexford**			
	Control kg ha^{-1} yr^{-1}	**Drought kg ha^{-1} yr^{-1}**	**%CAB**	***P*-value**	**Control kg ha^{-1} yr^{-1}**	**Drought kg ha^{-1} yr^{-1}**	**% CAB**	***P*-value**
L. perenne	8,680 (±278)	8,632 (±354)	-1	0.860	9,571 (±951)	9,248 (±190)	-3	0.804
C. intybus	10,121 (±510)	8,891 (±592)	-12	0.003	8,227 (±203)	8,490 (±1380)	+3	0.943
T. repens	11,370 (±551)	10,671 (±558)	-6	0.090	9,051 (±612)	8,573 (±351)	-5	0.599
T. pratense	16,091 (±478)	15,448 (±758)	-4	0.240	14,218 (±691)	12,683 (±322)	-11	0.260
Av. monoculture	11,566	10,911			10,267	9,749		
Mixture	16,346 (±369)	14,814 (±838)	-9	0.014	13,187 (±250)	12,537 (±202)	-5	0.604
Mixture benefit	4,780	3,903			2,920	2,788		

We found a relatively rapid recovery when soil water supply increased after the drought, even after the extreme drought stress at the site in Wexford. Such post-drought reaction might be explained in that we applied N fertilisers in equal amounts to both the rainfed control and the drought treatment. During the drought, much of this N was not available, and the recovery of post-drought yields was likely boosted by the increased availability of this N in the communities subjected to the drought treatment.

Conclusions

Aggregated across pre-drought, drought and post-drought harvests, the annual yield of temperate forage grassland species was largely resistant to a nine-week experimental summer drought. Multi-species mixtures showed strong overyielding (33%) of total annual yield under both rainfed control and drought treatment, in comparison to a 5% reduction in annual yield of the monocultures due to drought. Over short durations, the yield advantage from mixing species could compensate for yields being strongly affected by drought. Over the whole year, the strong overyielding of mixtures resulted in them exceeding the average yield of the rainfed monocultures under severe drought conditions. These results illustrate the high potential for multi-species mixtures to compensate for drought-induced yield losses.

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